

**WEST Search History**

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updated  
DIAGNOSTIC  
Searches  
6/04  
VSP

DATE: Tuesday, June 22, 2004

Hide?	Set Name	Query	Hit Count
<i>DB=USPT; PLUR=YES; OP=AND</i>			
<input type="checkbox"/>	L1	clostrid\$.ti same promoter\$.ti.	0
<input type="checkbox"/>	L2	clostrid\$.ti same transcript\$.ti.	0
<input type="checkbox"/>	L3	clostrid\$ near5 transcript\$	6
<i>DB=PGPB,USPT,USOC,EPAB,JPAB,DWPI,TDBD; PLUR=YES; OP=AND</i>			
<input type="checkbox"/>	L4	clostrid\$ near5 transcript\$	7
<input type="checkbox"/>	L5	L4 not l3	1

END OF SEARCH HISTORY

[Generate Collection](#)[Print](#)**Search Results - Record(s) 1 through 6 of 6 returned.**

- 
- ☐ 1. 6605431. 17 Aug 99; 12 Aug 03. Promoter elements and methods of use. Gourse; Richard L., et al. 435/6; 435/207 435/91.2 536/23.1. C12Q001/68 C12P019/34 C12N009/38 C07H021/04.
- 
- ☐ 2. 5955368. 06 Apr 98; 21 Sep 99. Expression system for clostridium species. Johnson; Eric A., et al. 435/488; 435/252.3 435/320.1 435/476 536/23.1 536/24.1. C12N001/21 C12N015/70 C12N015/74 C12N015/64.
- 
- ☐ 3. 5759845. 31 Jan 96; 02 Jun 98. Secretion of clostridium cellulase by E. coli. Yu; Ida Kuo. 435/277; 435/267 435/274. C12S003/02 C12S003/04.
- 
- ☐ 4. 5496725. 11 Aug 93; 05 Mar 96. Secretion of Clostridium cellulase by E. coli. Yu; Ida K.. 435/252.3; 435/209 435/252.33 435/254.11 435/320.1. C12N001/15 C12N001/21 C12N005/10 C12N009/42.
- 
- ☐ 5. 5418157. 22 Dec 92; 23 May 95. Recombinant 68,000 dalton collagenase of Clostridium histolyticum. Lin; Hun-Chi, et al. 435/220; 424/94.67 435/219 435/252.3 435/252.33 435/273 435/320.1 435/69.1 536/23.2 536/23.7. C12N009/52 C12N015/57 C12N015/70 C12N015/74.
- 
- ☐ 6. 5177017. 22 Mar 90; 05 Jan 93. Molecular cloning of the genes responsible for collagenase production from Clostridium histolyticum. Lin; Hun-Chi, et al. 435/252.33; 435/220 435/320.1 536/23.7. C12N015/57 C12N015/70 C12N015/31.
- 

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Terms	Documents
clostrid\$ near5 transcript\$	6

[Prev Page](#)[Next Page](#)[Go to Doc#](#)

File 155:MEDLINE(R) 1966-2004/Jun W2

(c) format only 2004 The Dialog Corp.

**\*File 155: Medline has been reloaded. Accession numbers**  
have changed. Please see HELP NEWS 154 for details.

Set	Items	Description
---	-----	-----
?s transcript? (3n)		promoter?
	316299	TRANSCRIPT?
	107776	PROMOTER?
S1	12190	TRANSCRIPT? (3N) PROMOTER?
?s clostrid? or perfring?		
	20130	CLOSTRID?
	5465	PERFRING?
S2	20223	CLOSTRID? OR PERFRING?
?s s1 and s2		
	12190	S1
	20223	S2
S3	22	S1 AND S2
?s s3/1998:2004		
	22	S3
	3229634	PY=1998 : PY=2004
S4	12	S3/1998:2004
?s s3 not s4		
	22	S3
	12	S4
S5	10	S3 NOT S4

Set Items Description

--- -----

Added File(s): 5, 34, 35, 48, 65, 71, 73, 91, 94, 98, 135, 144,  
149, 156, 159, 162, 164, 172, 266, 369, 370, 399, 434, 444,  
467

Previous sets have been retained; enter DISPLAY SETS to view them.

?repeat

Processing

Processed 20 of 26 files ...

Completed processing all files

2053476 TRANSCRIPT?

786872 PROMOTER?

S1 107558 TRANSCRIPT? (3N) PROMOTER?

140345 CLOSTRID?

30679 PERFRING?

S2 141097 CLOSTRID? OR PERFRING?

107558 S1

141097 S2

S3 555 S1 AND S2

Processing

Processed 10 of 26 files ...

>>>One or more prefixes are unsupported

>>> or undefined in one or more files.

>>>Year ranges not supported in one or more files

Completed processing all files

552 S3

32258795 PY=1998 : PY=2004

S4 120 S3/1998:2004

555 S3

120 S4

S5 435 S3 NOT S4

?rd

...examined 50 records (50)

...examined 50 records (100)

>>>Record 266:277084 ignored; incomplete bibliographic data, not retained -  
in RD set

>>>Record 266:215574 ignored; incomplete bibliographic data, not retained -  
in RD set

...examined 50 records (150)

...examined 50 records (200)

...examined 50 records (250)

...examined 50 records (300)

...examined 50 records (350)

...examined 50 records (400)

...completed examining records

S6 388 RD (unique items)

?s s6 and (nucleic? or plasmid? or heterolog? or nucleotid? or dna or cdna or mrna or r  
na or genetic)

Processing

Processed 10 of 26 files ...

Completed processing all files

388 S6

952906 NUCLEIC?

536092 PLASMID?

194134 HETEROLOG?

1537125 NUCLEOTID?

4611526 DNA

717004 CDNA

1171690 MRNA

2464046 RNA

3252297 GENETIC

S7 380 S6 AND (NUCLEIC? OR PLASMID? OR HETEROLOG? OR NUCLEOTID?  
OR DNA OR CDNA OR MRNA OR RNA OR GENETIC)

?s s6 and perfring?

388 S6

30679 PERFRING?  
S8 24 S6 AND PERFRING?  
?s s8 and s7

24 S8  
380 S7

S9 22 S8 AND S7

?target s9/all

Your TARGET search request will retrieve up to 50 of the statistically most relevant records.

Searching ALL records

...Processed 10 out of 26 files

...Processed 20 out of 26 files

...Processing Complete

S10 22 TARGET - S9

Ending TARGET search. Enter TARGET to do another search in the present file(s), or BEGIN new file(s). Enter LOGOFF to disconnect from Dialog

?t s10/6/all

10/6/1 (Item 1 from file: 155)  
13159543 PMID: 8828224

An upstream activating sequence containing curved DNA involved in activation of the *Clostridium perfringens* plc promoter.  
Sep 1996

10/6/2 (Item 2 from file: 399)  
DIALOG(R)File 399:(c) 2004 American Chemical Society. All rts. reserv.

The construction of a reporter system and use for the investigation of *Clostridium perfringens* gene expression

10/6/3 (Item 3 from file: 144)  
09115301 PASCAL No.: 90-0283682

Gene cloning shows the alpha-toxin of *Clostridium perfringens* to contain both sphingomyelinase and lecithinase activities  
1989

10/6/4 (Item 4 from file: 155)  
09432452 PMID: 1522810

Role of the upstream region containing an intrinsic DNA curvature in the negative regulation of the phospholipase C gene of *Clostridium perfringens*  
1992

10/6/5 (Item 5 from file: 34)  
03084944 Genuine Article#: NB994 Number of References: 32  
Title: THE VIRR GENE, A MEMBER OF A CLASS OF 2-COMPONENT RESPONSE REGULATORS, REGULATES THE PRODUCTION OF PERFRINGOLYSIN -O, COLLAGENASE, AND HEMAGGLUTININ IN *CLOSTRIDIUM* - PERFRINGENS (Abstract Available)

10/6/6 (Item 6 from file: 35)  
01601066 ORDER NO: NOT AVAILABLE FROM UNIVERSITY MICROFILMS INT'L.  
DEVELOPMENT OF A NOVEL EXPRESSION SYSTEM IN *CLOSTRIDIUM* PERFRINGENS (GENE EXPRESSION, SHUTTLE VECTOR)  
Year: 1997

10/6/7 (Item 7 from file: 144)  
08602368 PASCAL No.: 89-0151446  
Identification and molecular genetic analysis of replication functions of the bacteriocinogenic plasmid pIP404 from *Clostridium perfringens*  
1988

First Hit   Fwd Refs

L3: Entry 1 of 6

File: USPT

Aug 12, 2003

US-PAT-NO: 6605431

DOCUMENT-IDENTIFIER: US 6605431 B1

**\*\* See image for Certificate of Correction \*\***

TITLE: Promoter elements and methods of use

DATE-ISSUED: August 12, 2003

## INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Gourse; Richard L.	Madison	WI		
Estrem; Shawn T.	Greenwood	IN		
Ross; Wilma E.	Madison	WI		
Gaal; Tamas	Madison	WI		

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Wisconsin Alumni Research Foundation	Madison	WI			02

APPL-NO: 09/ 375673   [PALM]

DATE FILED: August 17, 1999

INT-CL: [07] C12 Q 1/68, C12 P 19/34, C12 N 9/38, C07 H 21/04

US-CL-ISSUED: 435/6; 435/91.2, 435/207, 536/23.1

US-CL-CURRENT: 435/6; 435/207, 435/91.2, 536/23.1

FIELD-OF-SEARCH: 435/6, 435/207, 435/91.2, 536/23.1

PRIOR-ART-DISCLOSED:

## U.S. PATENT DOCUMENTS

Search Selected

Search ALL

Clear

PAT-NO

ISSUE-DATE

PATENTEE-NAME

US-CL

☐ 6111077

August 2000

Sonenberg et al.

530/350

## OTHER PUBLICATIONS

Abstract of NIH Grant No. R01GM37048-01-03 for project entitled "Mechanism, Activation, and Control of rRNA Transcription," (1985).

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ART-UNIT: 1656

PRIMARY-EXAMINER: Benzion; Gary

ASSISTANT-EXAMINER: Tung; Joyce



ATTY-AGENT-FIRM: Muetting, Raasch & Gebhardt, P.A.

ABSTRACT:

The present invention provides novel polynucleotides that include promoter elements. elements. The present invention also provides methods and kits for identification of of compounds that alter transcription, preferably decrease transcription, of a polynucleotide. Also provided by the present invention are methods directed to producing RNA polynucleotides and polypeptides.

12 Claims, 14 Drawing figures

SYSTEM:OS - DIALOG OneSearch

File 155:MEDLINE(R) 1966-2004/Jun W2

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\*File 155: Medline has been reloaded. Accession numbers have changed. Please see HELP NEWS 154 for details.

File 5:Biosis Previews(R) 1969-2004/Jun W2

(c) 2004 BIOSIS

File 34:SciSearch(R) Cited Ref Sci 1990-2004/Jun W2

(c) 2004 Inst for Sci Info

File 35:Dissertation Abs Online 1861-2004/May

(c) 2004 ProQuest Info&Learning

File 48:SPORTDiscus 1962-2004/Jun

(c) 2004 Sport Information Resource Centre

File 65:Inside Conferences 1993-2004/Jun W3

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File 71:ELSEVIER BIOBASE 1994-2004/Jun W2

(c) 2004 Elsevier Science B.V.

File 73:EMBASE 1974-2004/Jun W2

(c) 2004 Elsevier Science B.V.

File 91:MANTIS(TM) 1880-2004/Jul

2001 (c) Action Potential

File 94:JICST-EPlus 1985-2004/May W5

(c)2004 Japan Science and Tech Corp(JST)

File 98:General Sci Abs/Full-Text 1984-2004/Jun

(c) 2004 The HW Wilson Co.

File 135:NewsRx Weekly Reports 1995-2004/Jun W1

(c) 2004 NewsRx

\*File 135: New newsletters are now added. See Help News135 for the complete list of newsletters.

File 144:Pascal 1973-2004/Jun W2

(c) 2004 INIST/CNRS

File 149:TGG Health&Wellness DB(SM) 1976-2004/Jun W2

(c) 2004 The Gale Group

File 156:ToxFile 1965-2004/May W5

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\*File 156: ToxFile now reloaded with 2004 MeSH.

Enter Help News156 for more information.

File 159:Cancerlit 1975-2002/Oct

(c) format only 2002 Dialog Corporation

\*File 159: Cancerlit ceases updating with immediate effect.

Please see HELP NEWS.

File 162:Global Health 1983-2004/May

(c) 2004 CAB International

File 164:Allied & Complementary Medicine 1984-2004/May

(c) 2004 BLHCIS

File 172:EMBASE Alert 2004/Jun W2

(c) 2004 Elsevier Science B.V.

File 266:FEDRIP 2004/Apr

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File 369:New Scientist 1994-2004/Jun W2

(c) 2004 Reed Business Information Ltd.

File 370:Science 1996-1999/Jul W3

(c) 1999 AAAS

\*File 370: This file is closed (no updates). Use File 47 for more current information.

File 399:CA SEARCH(R) 1967-2004/UD=14026

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Alert feature enhanced for multiple files, etc. See HELP ALERT.

File 434:SciSearch(R) Cited Ref Sci 1974-1989/Dec

(c) 1998 Inst for Sci Info

File 444:New England Journal of Med. 1985-2004/Jun W3

(c) 2004 Mass. Med. Soc.

File 467:ExtraMED(tm) 2000/Dec

(c) 2001 Informania Ltd.

\*File 467: For information about updating status please see Help News467.

\$0.34      0.063 DialUnits File135  
 \$0.34 Estimated cost File135  
          \$1.31      0.373 DialUnits File144  
          \$0.00    4 Type(s) in Format    6  
          \$0.00    4 Types  
 \$1.31 Estimated cost File144  
          \$0.68      0.154 DialUnits File149  
 \$0.68 Estimated cost File149  
          \$1.00      0.187 DialUnits File156  
          \$0.00    1 Type(s) in Format    6  
          \$0.00    1 Types  
 \$1.00 Estimated cost File156  
          \$0.52      0.175 DialUnits File159  
 \$0.52 Estimated cost File159  
          \$0.47      0.104 DialUnits File162  
 \$0.47 Estimated cost File162  
          \$0.22      0.062 DialUnits File164  
 \$0.22 Estimated cost File164  
          \$0.74      0.076 DialUnits File172  
 \$0.74 Estimated cost File172  
          \$0.27      0.079 DialUnits File266  
 \$0.27 Estimated cost File266  
          \$0.20      0.056 DialUnits File369  
 \$0.20 Estimated cost File369  
          \$0.32      0.092 DialUnits File370  
 \$0.32 Estimated cost File370  
          \$8.74      0.696 DialUnits File399  
          \$1.10    2 Type(s) in Format    6  
          \$1.10    2 Types  
 \$9.84 Estimated cost File399  
          \$8.14      0.397 DialUnits File434  
          \$0.00    2 Type(s) in Format    6  
          \$0.00    2 Types  
 \$8.14 Estimated cost File434  
          \$0.29      0.060 DialUnits File444  
 \$0.29 Estimated cost File444  
          \$0.31      0.048 DialUnits File467  
 \$0.31 Estimated cost File467  
          OneSearch, 26 files, 5.766 DialUnits FileOS  
 \$0.99 TELNET  
 \$52.51 Estimated cost this search  
 \$57.77 Estimated total session cost    6.826 DialUnits

### Status: Signed Off. (4 minutes)

### Status: Path 1 of [Dialog Information Services via Modem]

### Status: Initializing TCP/IP using (UseTelnetProto 1 ServiceID pto-dialog)  
 Trying 31060000009999...Open

DIALOG INFORMATION SERVICES

PLEASE LOGON:

\*\*\*\*\* HHHHHHHH SSSSSSSS?

### Status: Signing onto Dialog

\*\*\*\*\*

ENTER PASSWORD:

\*\*\*\*\* HHHHHHHH SSSSSSSS? \*\*\*\*\*

Welcome to DIALOG

### Status: Connected

Dialog level 04.10.00D

Reconnected in file OS 22jun04 14:09:53

\* \* \* \*

10/6/8 (Item 8 from file: 5)  
0006699658 BIOSIS NO.: 198988014773  
NUCLEOTIDE SEQUENCE ANALYSIS AND EXPRESSION STUDIES OF A CHLORAMPHENICOL  
ACETYLTRANSFERASE-CODING GENE FROM CLOSTRIDIUM-PERFRINGENS  
1989

10/6/9 (Item 9 from file: 155)  
12832817 PMID: 8566714  
Transcriptional analysis of the beta-galactosidase gene (pbq) in  
Clostridium perfringens.  
Nov 1 1995

10/6/10 (Item 10 from file: 144)  
09302147 PASCAL No.: 91-0092521  
Cloning and sequencing of the genes encoding acid-soluble spore proteins  
from Clostridium perfringens  
1990

10/6/11 (Item 11 from file: 399)  
DIALOG(R) File 399:(c) 2004 American Chemical Society. All rts. reserv.

Comparison of the alpha-toxin genes of Clostridium perfringens type A and  
C strains: Evidence for extragenic regulation of transcription

10/6/12 (Item 12 from file: 34)  
03238985 Genuine Article#: NP484 Number of References: 80  
Title: IDENTIFICATION AND MOLECULAR ANALYSIS OF A LOCUS THAT REGULATES  
EXTRACELLULAR TOXIN PRODUCTION IN CLOSTRIDIUM - PERFRINGENS (  
Abstract Available)

10/6/13 (Item 13 from file: 34)  
01195255 Genuine Article#: GD031 Number of References: 29  
Title: CLONING, MAPPING, AND MOLECULAR CHARACTERIZATION OF THE RIBOSOMAL-  
RNA OPERONS OF CLOSTRIDIUM - PERFRINGENS (Abstract Available)

10/6/14 (Item 14 from file: 34)  
00847254 Genuine Article#: FA766 Number of References: 40  
Title: RELATIONSHIP BETWEEN THE CLOSTRIDIUM - PERFRINGENS CATQ  
GENE-PRODUCT AND CHLORAMPHENICOL ACETYLTRANSFERASES FROM OTHER BACTERIA  
(Abstract Available)

10/6/15 (Item 15 from file: 434)  
09269425 Genuine Article#: R9221 Number of References: 50  
Title: MOLECULAR-CLONING AND NUCLEOTIDE -SEQUENCE OF THE ALPHA-TOXIN  
(PHOSPHOLIPASE-C) OF CLOSTRIDIUM - PERFRINGENS

10/6/16 (Item 16 from file: 156)  
00582052 NLM Doc No: CRISP/98/AI27655-10 Sec. Source ID:  
CRISP/98/AI27655-10  
LISTERIA HEMOLYSIN AND ESCAPE FROM A VACUOLE  
1997

10/6/17 (Item 17 from file: 5)  
0007865675 BIOSIS NO.: 199192111446  
CLONING MAPPING AND MOLECULAR CHARACTERIZATION OF THE RNA OPERONS OF  
CLOSTRIDIUM -PERFRINGENS

1991

10/6/18 (Item 18 from file: 144)

08706627 PASCAL No.: 89-0255883

Studies of UV-inducible promoters from *Clostridium perfringens* in vivo  
and in vitro  
1988

10/6/19 (Item 19 from file: 434)

09379536 Genuine Article#: T7870 Number of References: 48

Title: PHOSPHOLIPASE-C AND HEMOLYTIC ACTIVITIES OF *CLOSTRIDIUM* -  
*PERFRINGENS* ALPHA-TOXIN CLONED IN *ESCHERICHIA-COLI* - SEQUENCE AND  
HOMOLOGY WITH A *BACILLUS-CEREUS* PHOSPHOLIPASE-C

10/6/20 (Item 20 from file: 34)

03302660 Genuine Article#: NU575 Number of References: 51

Title: ORGANIZATION OF THE BOTULINUM NEUROTOXIN C1 GENE AND ITS ASSOCIATED  
NONTXIC PROTEIN GENES IN *CLOSTRIDIUM-BOTULINUM-C-468* (Abstract  
Available)

10/6/21 (Item 21 from file: 155)

09142587 PMID: 1309513

Nucleotide sequence of the lecithinase operon of *Listeria monocytogenes*  
and possible role of lecithinase in cell-to-cell spread.  
Jan 1992

10/6/22 (Item 22 from file: 34)

01712426 Genuine Article#: HV090 Number of References: 39

Title: PURIFICATION AND CHARACTERIZATION OF AN ADP-RIBOSYLTRANSFERASE  
PRODUCED BY *CLOSTRIDIUM-LIMOSUM* (Abstract Available)

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\$1.45 Estimated cost File155

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\$0.00 2 Types

\$3.72 Estimated cost File5

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\$0.00 6 Type(s) in Format 6

\$0.00 6 Types

\$13.19 Estimated cost File34

\$0.49 0.119 DialUnits File35

\$0.00 1 Type(s) in Format 6

\$0.00 1 Types

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\$0.46 0.086 DialUnits File48

\$0.46 Estimated cost File48

\$0.88 0.236 DialUnits File65

\$0.88 Estimated cost File65

\$1.64 0.207 DialUnits File71

\$1.64 Estimated cost File71

\$3.97 0.405 DialUnits File73

\$3.97 Estimated cost File73

\$0.27 0.063 DialUnits File91

\$0.27 Estimated cost File91

\$0.54 0.156 DialUnits File94

\$0.54 Estimated cost File94

\$0.26 0.110 DialUnits File98

\$0.26 Estimated cost File98

DIALOG(R) File 155:MEDLINE(R)  
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13159543 PMID: 8828224

**An upstream activating sequence containing curved DNA involved in activation of the *Clostridium perfringens* plc promoter.**

Matsushita C; Matsushita O; Katayama S; Minami J; Takai K; Okabe A

Department of Microbiology, Kagawa Medical School, Japan.

Microbiology (Reading, England) (ENGLAND) Sep 1996, 142 ( Pt 9)  
p2561-6, ISSN 1350-0872 Journal Code: 9430468

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Subfile: INDEX MEDICUS

The plc gene, which encodes phospholipase C (alpha-toxin) of *Clostridium perfringens*, possesses three poly(A) tracts forming an intrinsically curved DNA region immediately upstream of the promoter. The in vivo transcriptional activity of the plasmid-borne plc gene was stimulated by this curved-DNA-containing sequence, depending on its proper linear and rotational orientation. The in vitro transcriptional activity of the plc gene was also stimulated by the upstream sequence. In addition, the stimulatory effect of the sequence and the degree of DNA bending were greater at lower temperature, as was demonstrated by both in vitro and in vivo transcription assays, and a gel-mobility assay, respectively. A similar temperature effect was also observed with the chromosomal plc gene. These observations suggest that the upstream DNA curvature per se stimulates the initiation of transcription of the plc gene, possibly through direct contact with RNA polymerase.

Tags: Support, Non-U.S. Gov't

Descriptors: *Clostridium perfringens* --genetics--GE; \*Phospholipase C --genetics--GE; Base Sequence; Chromosome Mapping; Chromosomes--genetics --GE; Chromosomes--physiology--PH; DNA--physiology--PH; Gene Expression Regulation, Bacterial; Molecular Sequence Data; Mutagenesis, Insertional; Mutagenesis, Site-Directed; Nucleic Acid Conformation; Plasmids--genetics --GE; Plasmids--physiology--PH; Promoter Regions (Genetics); Sequence Deletion; Temperature; Transcription, Genetic

CAS Registry No.: 0 (Plasmids); 9007-49-2 (DNA)

Enzyme No.: EC 3.1.4.3 (Phospholipase C)

Record Date Created: 19970113

Record Date Completed: 19970113

5/9/2

DIALOG(R) File 155:MEDLINE(R)  
(c) format only 2004 The Dialog Corp. All rts. reserv.

12832817 PMID: 8566714

**Transcriptional analysis of the beta-galactosidase gene (pbg) in *Clostridium perfringens*.**

Kobayashi T; Shimizu T; Hayashi H

Department of Microbiology, University of Tsukuba, Ibaraki, Japan.

FEMS microbiology letters (NETHERLANDS) Nov 1 1995, 133 (1-2) p65-9,  
ISSN 0378-1097 Journal Code: 7705721

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Subfile: INDEX MEDICUS

The mode of expression of the beta-galactosidase gene (pbg) of *Clostridium perfringens* was examined. The pbg gene was transcribed on a single 3.7-kb mRNA. The transcript contained a message for ORF54, located upstream of the pbg gene in the chromosome, indicating that ORF54 and the pbg gene comprise one operon (pbg operon). Expression of the pbg operon was induced by lactose at the transcriptional level. The promoter structure of the pbg operon was characterized by many palindrome structures and direct repeats, which suggests that there might be some catabolite

regulation of the expression of the pbj operon in *C. perfringens* .

Tags: Support, Non-U.S. Gov't

Descriptors: **Clostridium perfringens** --genetics--GE; \*Genes, Bacterial  
--genetics--GE; \*Transcription, Genetic--genetics--GE; \*beta-Galactosidase  
--genetics--GE; Base Sequence; Chromosome Mapping; Cloning, Molecular;  
**Clostridium perfringens** --enzymology--EN; Lactose--metabolism--ME;  
Molecular Sequence Data; RNA, Bacterial--analysis--AN; RNA, Messenger  
--analysis--AN

CAS Registry No.: 0 (RNA, Bacterial); 0 (RNA, Messenger); 63-42-3  
(Lactose)

Enzyme No.: EC 3.2.1.23 (beta-Galactosidase)

Record Date Created: 19960301

Record Date Completed: 19960301

5/9/3

DIALOG(R) File 155:MEDLINE(R)

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10261492 PMID: 7961408

**Sporulation and primary sigma factor homologous genes in *Clostridium acetobutylicum*.**

Sauer U; Treuner A; Buchholz M; Santangelo J D; Durre P

Institut fur Mikrobiologie, Georg-August-Universitat Gottingen, Germany.

Journal of bacteriology (UNITED STATES) Nov 1994, 176 (21) p6572-82,

ISSN 0021-9193 Journal Code: 2985120R

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Subfile: INDEX MEDICUS

Using a PCR-based approach, we have cloned various sigma factor homologous genes from ***Clostridium acetobutylicum*** DSM 792. The nucleotide sequence of the dnaE-sigA operon has been determined and predicts two genes encoding 69- and 43-kDa proteins. The deduced DnaE amino acid sequence has approximately 30% amino acid identity with protein sequences of other primases. The putative sigA gene product shows high homology to primary sigma factors of various bacteria, most significantly to *Bacillus subtilis* and *Staphylococcus aureus*. Northern (RNA) blot analysis revealed that both genes from an operon, which is clearly expressed under conditions that allow for cell division. A promoter sequence with significant homology to the sigma H-dependent *Bacillus* **promoters** preceded the determined **transcriptional** start point, 182 bp upstream of the GUG start codon of dnaE. The homologous genes to *Bacillus* spp. sporulation sigma factors G, E, and K have been cloned and sequenced. Indirect evidence for the existence of sigma F was obtained by identification of a DNA sequence homologous to the respective *Bacillus* consensus promoter. Southern hybridization analysis indicated the presence of sigma D and sigma H homologous genes in *C. acetobutylicum*. A new gene group conserved within the eubacteria, but with yet unspecified functions, is described. The data presented here provide strong evidence that at least some of the complex regulation features of sporulation in *B. subtilis* are conserved in *C. acetobutylicum* and possibly ***Clostridium* spp.**

Tags: Comparative Study; Support, Non-U.S. Gov't

Descriptors: **Clostridium** --genetics--GE; \*Genes, Bacterial--genetics--GE  
; \*Sigma Factor--genetics--GE; \*Spores, Bacterial--genetics--GE; Amino Acid  
Sequence; Bacterial Proteins--genetics--GE; Base Sequence; Cloning,  
Molecular; **Clostridium** --growth and development--GD; DNA Polymerase III  
--genetics--GE; DNA-Directed RNA Polymerases--genetics--GE; Gene Expression  
Regulation, Bacterial; Genomic Library; Molecular Sequence Data; Operon  
--genetics--GE; Polymerase Chain Reaction; Sequence Analysis, DNA; Sequence  
Homology, Amino Acid; Spores, Bacterial--growth and development--GD;  
Transcription Factors--genetics--GE; Transcription, Genetic

Molecular Sequence Databank No.: GENBANK/L23317; GENBANK/Z23079;  
GENBANK/Z23080

CAS Registry No.: 0 (Bacterial Proteins); 0 (DnaE protein); 0 (Sigma  
Factor); 0 (Transcription Factors); 0 (sigma K); 0 (sigma-E factor)

Enzyme No.: EC 2.7.7.- (DNA Polymerase III); EC 2.7.7.- (RNA polymerase sigma 70); EC 2.7.7.- (RNA polymerase sigma G); EC 2.7.7.6 (DNA-Directed RNA Polymerases)  
Gene Symbol: dnaE; sigA; sigE; sigG; sigK  
Record Date Created: 19941130  
Record Date Completed: 19941130

5/9/4

DIALOG(R) File 155:MEDLINE(R)

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09432452 PMID: 1522810

**Role of the upstream region containing an intrinsic DNA curvature in the negative regulation of the phospholipase C gene of Clostridium perfringens .**

Toyonaga T; Matsushita O; Katayama S; Minami J; Okabe A  
Department of Microbiology, Kagawa Medical School, Japan.  
Microbiology and immunology (JAPAN) 1992, 36 (6) p603-13, ISSN 0385-5600 Journal Code: 7703966  
Document type: Journal Article  
Languages: ENGLISH  
Main Citation Owner: NLM  
Record type: Completed  
Subfile: INDEX MEDICUS

The phospholipase C (alpha-toxin) gene (plc) of **Clostridium perfringens** was cloned into pUC19 and the effects of the upstream regions on expression of the plc gene were examined in Escherichia coli JM109. When the 0.7-kb region just upstream of the putative -35 site of the gene was deleted, production of phospholipase C increased approximately 10-fold. Northern blot hybridization analysis of the plc transcript showed that the upstream region inhibited **transcription** from the plc **promoter** . Nucleotide sequencing of this upstream region revealed that there are three periodically repeated (dA)5-6 tracts between positions -66 and -40 of the plc gene. A fragment containing this sequence showed anomalously slow electrophoretic mobility at low temperature, indicating that the region immediately upstream of the plc promoter is a locus of sequence directed DNA-bending. Nested deletions of the upstream region were created from its 5' end by exonuclease III and the effects of deletions on the expression of the plc gene were examined. When the 77-bp fragment containing the two (dA)5-6 tracts were deleted, phospholipase C production increased markedly. These results indicate that the intrinsic DNA curvature upstream of the plc promoter is involved in the negative regulation of the plc gene transcription.

Tags: Comparative Study; Support, Non-U.S. Gov't

Descriptors: Bacterial Toxins--genetics--GE; \* **Clostridium perfringens** --genetics--GE; \*DNA, Bacterial--genetics--GE; \*Gene Expression Regulation, Enzymologic; \*Genes, Bacterial--genetics--GE; \*Phospholipase C--genetics--GE; Base Sequence; Blotting, Northern; Chromosome Deletion; **Clostridium perfringens** --enzymology--EN; Escherichia coli--genetics--GE; Molecular Sequence Data; Phospholipase C--metabolism--ME; Plasmids--genetics--GE; RNA, Messenger--metabolism--ME; Restriction Mapping; Transfection

CAS Registry No.: 0 (Bacterial Toxins); 0 (DNA, Bacterial); 0 (Plasmids); 0 (RNA, Messenger)

Enzyme No.: EC 3.1.4.- (**Clostridium perfringens** alpha-toxin); EC 3.1.4.3 (Phospholipase C)

Record Date Created: 19921015

Record Date Completed: 19921015

5/9/5

DIALOG(R) File 155:MEDLINE(R)

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09287605 PMID: 1349602

**Cloning, sequencing, and molecular analysis of the groESL operon of Clostridium acetobutylicum.**



Narberhaus F; Bahl H  
Institut fur Mikrobiologie, Georg-August-Universitat Gottingen, Germany.  
Journal of bacteriology (UNITED STATES) May 1992, 174 (10) p3282-9,  
ISSN 0021-9193 Journal Code: 2985120R  
Document type: Journal Article  
Languages: ENGLISH  
Main Citation Owner: NLM  
Record type: Completed  
Subfile: INDEX MEDICUS

The groESL operon of *Clostridium acetobutylicum* was cloned in *Escherichia coli* by using a gene probe of *E. coli* groESL. Sequencing of a positively reacting 2.2-kbp HindIII fragment contained in the recombinant plasmid pFN1 and a 2.5-kbp XbaI fragment present in pFN4 revealed that both fragments partially overlapped and together spanned 3,493 bp of the *clostridial* chromosome. Two complete open reading frames (288 and 1632 bp) were found and identified as the groES- and groEL-homologous genes of *C. acetobutylicum*, respectively. The 3' end of a third gene (orfZ), which was divergently transcribed, showed no significant homology to other sequences available in the EMBL and GenBank data bases. The length of the groESL-specific mRNA (2.2 kb), a transcription terminator downstream of groEL, and a transcription start site upstream of groES, identified by primer extension analysis, indicated that groES and groEL of *C. acetobutylicum* are organized in a bicistronic operon. From the transcription start site, the promoter structure 5'-TTGCTA (17 bp) TATTAT that shows high homology to the consensus promoter sequence of gram-positive bacteria as well as *E. coli* was deduced. Transcription of the groESL operon was strongly heat inducible, and maximum levels of mRNA were detected 15 min after heat shock from 30 to 42 degrees C. An 11-bp inverted repeat, located between promoter and translation start sites of groES and partially identical with similar structures in front of several heat shock genes of other bacteria, may play an important role in the regulation of heat shock gene expression in this organism.

Tags: Comparative Study; Support, Non-U.S. Gov't  
Descriptors: *Clostridium* --genetics--GE; \*Heat-Shock Proteins--genetics--GE; \*RNA, Messenger--analysis--AN; Amino Acid Sequence; Bacterial Proteins--genetics--GE; Base Sequence; Cloning, Molecular; Consensus Sequence; *Escherichia coli*--genetics--GE; Gene Expression Regulation, Bacterial; GroEL Protein; GroES Protein; Molecular Sequence Data; Nucleic Acid Conformation; Operon--genetics--GE; Regulatory Sequences, Nucleic Acid--genetics--GE; Repetitive Sequences, Nucleic Acid--genetics--GE; Sequence Homology, Nucleic Acid; Transcription, Genetic  
Molecular Sequence Databank No.: GENBANK/M74572; GENBANK/M79367; GENBANK/M79368; GENBANK/M79369; GENBANK/M79370; GENBANK/M79371; GENBANK/M79372; GENBANK/M87491; GENBANK/M87492; GENBANK/M87836  
CAS Registry No.: 0 (Bacterial Proteins); 0 (GroEL Protein); 0 (GroES Protein); 0 (Heat-Shock Proteins); 0 (RNA, Messenger)  
Record Date Created: 19920609  
Record Date Completed: 19920609

5/9/6  
DIALOG(R) File 155:MEDLINE(R)  
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09192767 PMID: 1740123

Structure of the *Clostridium thermocellum* gene licB and the encoded beta-1,3-1,4-glucanase. A catalytic region homologous to *Bacillus lichenases* joined to the reiterated domain of clostridial cellulases.

Schimming S; Schwarz W H; Staudenbauer W L  
Institute for Microbiology, Technical University Munich, Federal Republic of Germany.

European journal of biochemistry / FEBS (GERMANY) Feb 15 1992, 204  
(1) p13-9, ISSN 0014-2956 Journal Code: 0107600  
Document type: Journal Article  
Languages: ENGLISH  
Main Citation Owner: NLM  
Record type: Completed

Subfile: INDEX MEDICUS

The nucleotide sequence of the **Clostridium** thermocellum gene licB, coding for a thermoactive beta-1,3-1,4-glucanase, has been determined. The gene is located downstream, but in opposite orientation to the beta-glucosidase gene bglA. A coding region of 1002 bp is flanked by canonical **promoter** and **transcription** terminator sequences. The primary translation product of the licB gene has a predicted molecular mass of 37,896 Da. The protein sequence can be divided into several discrete segments: an N-terminal signal peptide, a catalytic region, a segment rich in Pro and Thr residues and a C-terminal reiterated domain. The catalytic region shows close similarity to lichenases of bacilli (52-58% identity) and Fibrobacter succinogenes (35% identity), but is unrelated to barley beta-1,3-1,4-glucanases. It consists of two domains, which in the case of the F. succinogenes lichenase are arranged in reversed order to that of C. thermocellum and Bacillus lichenases. The C-terminal reiterated domain of C. thermocellum lichenase is homologous to the duplicated non-catalytic domain of endo-beta-1,4-glucanases and xylanase Z from the same organism. This domain is considered a characteristic feature of **clostridial** cellulases organized as multienzyme complex (cellulosome). The beta-1,3-1,4-glucanase encoded by the licB gene might therefore be an additional enzyme component of the C. thermocellum cellulosome.

Tags: Comparative Study; Support, Non-U.S. Gov't

Descriptors: **Clostridium** --genetics--GE; \*Genes, Bacterial; \*Glycoside Hydrolases--genetics--GE; Amino Acid Sequence; Base Sequence; Binding Sites; Cellulase--chemistry--CH; **Clostridium** --enzymology--EN; DNA, Bacterial --chemistry--CH; Glycoside Hydrolases--chemistry--CH; Molecular Sequence Data; Molecular Weight; Restriction Mapping; Sequence Homology, Nucleic Acid

Molecular Sequence Databank No.: GENBANK/M76990; GENBANK/X63355; GENBANK/X65174; GENBANK/X65175; GENBANK/X65176; GENBANK/X65177; GENBANK/X65178; GENBANK/X65179; GENBANK/X65180; GENBANK/X65181

CAS Registry No.: 0 (DNA, Bacterial)

Enzyme No.: EC 3.2.1. (Glycoside Hydrolases); EC 3.2.1.4 (Cellulase); EC 3.2.1.73 (licheninase)

Gene Symbol: licB

Record Date Created: 19920324

Record Date Completed: 19920324

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DIALOG(R) File 155:MEDLINE(R)

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09142587 PMID: 1309513

**Nucleotide sequence of the lecithinase operon of Listeria monocytogenes and possible role of lecithinase in cell-to-cell spread.**

Vazquez-Boland J A; Kocks C; Dramsi S; Ohayon H; Geoffroy C; Mengaud J; Cossart P

Unite de Genie Microbiologique, Institut Pasteur, Paris, France.

Infection and immunity (UNITED STATES) Jan 1992, 60 (1) p219-30, ISSN 0019-9567 Journal Code: 0246127

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Subfile: INDEX MEDICUS

The lecithinase gene of the intracellular pathogen Listeria monocytogenes, plcB, was identified in a 5,648-bp DNA fragment which expressed lecithinase activity when cloned into Escherichia coli. This fragment is located immediately downstream of the previously identified gene mpl (prtA). It contains five open reading frames, named actA, plcB, and ORFX, -Y, and -Z, which, together with mpl, form an operon, since a 5.7-kb-long **transcript** originates from a **promoter** located upstream of mpl (J. Mengaud, C. Geoffroy, and P. Cossart, Infect. Immun. 59:1043-1049, 1991). A second promoter was detected in front of actA which encodes a putative membrane protein containing a region of internal repeats. plcB encodes the lecithinase, a predicted 289-amino-acid protein homologous to

the phosphatidylcholine-specific phospholipases C of *Bacillus cereus* and *Clostridium perfringens* (alpha-toxin). *plcB* mutants produce only small plaques on fibroblast monolayers, and an electron microscopic analysis of infected macrophages suggests that lecithinase is involved in the lysis of the two-membrane vacuoles that surround the bacteria after cell-to-cell spread. On the opposite DNA strand, downstream of the operon, three more open reading frames, *ldh*, *ORFA*, and *ORFB*, were found. The deduced amino acid sequence of the first one is homologous to lactate dehydrogenases. Low-stringency Southern hybridization experiments suggest that these three open reading frames lie outside of the *L. monocytogenes* virulence region: *mpl* and *actA* were specific for *L. monocytogenes*, sequences hybridizing to *plcB* were detected in *L. ivanovii* and *L. seeligeri*, and sequences hybridizing to *ORFX*, *-Y*, and *-Z* were found in *L. innocua*. In contrast to this, sequences hybridizing to *ldh* or *ORFB* were detected in all *Listeria* species (including the nonpathogenic ones).

Tags: Comparative Study; In Vitro; Support, Non-U.S. Gov't

Descriptors: \**Listeria monocytogenes*--enzymology--EN; \*Operon--genetics--GE; \*Phospholipases--genetics--GE; Amino Acid Sequence; Animals; Bacterial Outer Membrane Proteins--genetics--GE; Base Sequence; Blotting, Southern; Cloning, Molecular; DNA Transposable Elements; *Listeria monocytogenes*--pathogenicity--PY; Mice; Microscopy, Electron; Molecular Sequence Data; Phospholipases--physiology--PH; Plaque Assay; Promoter Regions (Genetics)--genetics--GE; Restriction Mapping; Sequence Homology, Nucleic Acid; Virulence--genetics--GE

Molecular Sequence Databank No.: GENBANK/M63610; GENBANK/M63611; GENBANK/M63612; GENBANK/M63613; GENBANK/M63614; GENBANK/M63615; GENBANK/M63616; GENBANK/M63617; GENBANK/M82881; GENBANK/X63185

CAS Registry No.: 0 (Bacterial Outer Membrane Proteins); 0 (DNA Transposable Elements)

Enzyme No.: EC 3.1.- (Phospholipases)

Gene Symbol: *-y*; *-z*; *ORFX*; *actA*; *hyl*; *ldh*; *mpl*; *plcB*; *prtA*

Record Date Created: 19920212

Record Date Completed: 19920212

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DIALOG(R) File 155:MEDLINE(R)

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09024086 PMID: 1909624

**Structure of the beta-glucosidase gene *bglA* of *Clostridium thermocellum*. Sequence analysis reveals a superfamily of cellulases and beta-glycosidases including human lactase/phlorizin hydrolase.**

Grabnitz F; Seiss M; Rucknagel K P; Staudenbauer W L

Institute for Microbiology, Technical University Munich, Federal Republic of Germany.

European journal of biochemistry/ FEBS (GERMANY) Sep 1 1991, 200 (2) p301-9, ISSN 0014-2956 Journal Code: 0107600

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Subfile: INDEX MEDICUS

The nucleotide sequence of the *Clostridium thermocellum* gene *bglA*, coding for the thermostable beta-glucosidase A, has been determined. The coding region of 1344 bp was identified by comparison with the N-terminal amino acid sequence of recombinant beta-glucosidase A purified from *Escherichia coli*. The deduced amino acid sequence corresponds to a protein of 51,482 Da. The coding region is flanked by putative **promoter** and **transcription** terminator sequences. The protein is unrelated to beta-glucosidase B of *C. thermocellum*, but has a high level of similarity with other bacterial beta-glucosidases and phospho-beta-glucosidases. Similarity is also observed with the beta-galactosidase of the archaeobacterium *Sulfolobus solfataricus*. Unexpectedly, it was found that human lactase-phlorizin hydrolase contains three copies of a sequence closely related to *C. thermocellum* beta-glucosidase A (up to 40% sequence identity). These diverse beta-glucosidases can therefore be grouped into an

enzyme family (BGA) of common structural design. Sequence comparison by hydrophobic cluster analysis revealed that all BGA enzymes share a well conserved region which is homologous to the catalytic domain of the widely distributed cellulase family A. A distinctive feature of this domain is the sequence motif His-Asn-Glu-Pro in which the catalytic residues His and Glu are separated by 35-55 amino acid residues. The cellulase family A and the beta-glucosidase family BGA might thus be considered as members of a protein super-family comprising beta-glucanases and beta-glycosidases from all three primary kingdoms of living organisms.

Tags: Human; Support, Non-U.S. Gov't

Descriptors: Cellulase--genetics--GE; \* **Clostridium** --genetics--GE; \*Genes, Bacterial; \*Glycosylceramidase--genetics--GE; \*beta-Galactosidase --genetics--GE; \*beta-Glucosidase--genetics--GE; Amino Acid Sequence; Base Sequence; Electrophoresis, Polyacrylamide Gel; Lactase; Molecular Sequence Data; Multigene Family; Promoter Regions (Genetics); Restriction Mapping; Sequence Alignment; Sequence Homology, Nucleic Acid; Transcription, Genetic Molecular Sequence Databank No.: GENBANK/M60272; GENBANK/M60273; GENBANK/M60352; GENBANK/M60353; GENBANK/M60354; GENBANK/S52677; GENBANK/X56257; GENBANK/X57950; GENBANK/X57951; GENBANK/X60268

Enzyme No.: EC 3.2.1.108 (Lactase); EC 3.2.1.21 (beta-Glucosidase); EC 3.2.1.23 (beta-Galactosidase); EC 3.2.1.4 (Cellulase); EC 3.2.1.62 (Glycosylceramidase)

Gene Symbol: bglA

Record Date Created: 19911015

Record Date Completed: 19911015

5/9/9

DIALOG(R) File 155:MEDLINE(R)

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07187369 PMID: 3733758

**In vivo and in vitro transcription of the *Clostridium pasteurianum* ferredoxin gene. Evidence for "extended" promoter elements in gram-positive organisms.**

Graves M C; Rabinowitz J C

Journal of biological chemistry (UNITED STATES) Aug 25 1986, 261 (24)

pl1409-15, ISSN 0021-9258 Journal Code: 2985121R

Contract/Grant No.: AI6712; AI; NIAID; AM2109-28; AM; NIADDK

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Subfile: INDEX MEDICUS

Analysis of ***Clostridium pasteurianum*** genomic DNA indicates that the ferredoxin (Fd) gene is present in a single copy. The cloned Fd gene previously described (Graves, M.C., Mullenbach, G. T., and Rabinowitz, J. C. (1985) Proc. Natl. Acad. Sci. U. S. A. 82, 1653-1657) was used to map in vivo and in vitro synthesized Fd transcripts. The in vivo mRNA was sized in two ways: by Northern hybridization analysis, and more directly from the known DNA sequence after the 5'- and 3'-termini were identified. The 5'-end was determined by primer extension-dideoxy sequencing and the 3'-end by S1 nuclease mapping. The monocistronic Fd mRNA contains about 255 nucleotides and, thus, is one of the shortest bacterial mRNAs yet described. We also examined the Fd transcripts produced by *Escherichia coli* transformed with the plasmid containing the Fd gene. *E. coli* RNA polymerase most likely recognizes the same promoter (P1) as the **clostridial** polymerase, and furthermore, efficiently uses an additional promoter (P2) that is poorly recognized by the normal host enzyme. For comparison, in vitro transcripts were generated by *E. coli* and *Bacillus subtilis* RNA polymerases. In vitro, only promoter P1 is used by either *E. coli* or *B. subtilis* RNA polymerase. The 3'-end of each of the four types of transcripts occurs essentially at the same location and maps to within a large dyad symmetry element. Comparison of the Fd promoter with other Gram-positive promoters reveals that some sequences outside of the traditional Pribnow and -35 regions are conserved. This analysis indicates that an "extended" promoter recognition site may be required in these organisms.

Tags: Support, U.S. Gov't, P.H.S.  
Descriptors: **Clostridium** --genetics--GE; \*Ferredoxins--genetics--GE; \*  
**Promoter** Regions (Genetics); \* **Transcription**, Genetic; Base Sequence;  
Electrophoresis, Polyacrylamide Gel; Nucleic Acid Conformation; Nucleic  
Acid Hybridization; RNA, Messenger--metabolism--ME  
Molecular Sequence Databank No.: GENBANK/M11214; GENBANK/M13633;  
GENBANK/M13682  
CAS Registry No.: 0 (Ferredoxins); 0 (RNA, Messenger)  
Record Date Created: 19860919  
Record Date Completed: 19860919

5/9/10

DIALOG(R) File 155:MEDLINE(R)

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07159939 PMID: 3013834

**Cloning and expression in Escherichia coli of the gene for  
10-formyltetrahydrofolate synthetase from Clostridium acidurici ("**  
**Clostridium acidi-urici").**

Whitehead T R; Rabinowitz J C

Journal of bacteriology (UNITED STATES) Jul 1986, 167 (1) p205-9,

ISSN 0021-9193 Journal Code: 2985120R

Contract/Grant No.: AM02109; AM; NIADDK

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Subfile: INDEX MEDICUS

The gene for 10-formyltetrahydrofolate synthetase (EC 6.3.4.3) from the  
purinolytic anaerobic bacterium **Clostridium** acidurici ("**Clostridium**  
acidi-urici") was cloned into Escherichia coli JM83 with plasmid pUC8. A C.  
acidurici genomic library was prepared in E. coli from a partial Sau3A  
digest and screened with antibody against the synthetase. Of 10  
antibody-positive clones, 1 expressed a high level of synthetase activity.  
Sodium dodecyl sulfate-polyacrylamide gel electrophoresis and immunoblot  
analysis demonstrated that the protein synthesized in E. coli had the same  
subunit molecular weight as the C. acidurici enzyme. The gene was located  
on an 8.3-kilobase genomic insert and appeared to be transcribed from its  
own promoter. Analysis of genomic digests with a fragment of the synthetase  
gene indicated that one copy of the gene was present in the C. acidurici  
chromosome.

Tags: Support, Non-U.S. Gov't; Support, U.S. Gov't, P.H.S.

Descriptors: Cloning, Molecular; \* **Clostridium** --genetics--GE;  
\*Escherichia coli--genetics--GE; \*Formate-Tetrahydrofolate Ligase--genetics  
--GE; \*Ligases--genetics--GE; **Clostridium** --enzymology--EN; DNA  
Restriction Enzymes; Escherichia coli--enzymology--EN; Formate-Tetrahydrofo  
late Ligase--biosynthesis--BI; Genes, Bacterial; Nucleic Acid Hybridization  
; **Promoter** Regions (Genetics); **Transcription**, Genetic

Enzyme No.: EC 3.1.21 (DNA Restriction Enzymes); EC 6. (Ligases); EC  
6.3.4.3 (Formate-Tetrahydrofolate Ligase)

Record Date Created: 19860811

Record Date Completed: 19860811

?add medicine

22jun04 14:02:13 User228206 Session D2186.2

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\$2.10 10 Type(s) in Format 9

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\$5.00 Estimated cost File155

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\$5.24 Estimated cost this search

\$5.26 Estimated total session cost 1.059 DialUnits

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10/9/1 (Item 1 from file: 155)

DIALOG(R) File 155:MEDLINE(R)

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13159543 PMID: 8828224

**An upstream activating sequence containing curved DNA involved in activation of the *Clostridium perfringens* plc promoter.**

Matsushita C; Matsushita O; Katayama S; Minami J; Takai K; Okabe A

Department of Microbiology, Kagawa Medical School, Japan.

Microbiology (Reading, England) (ENGLAND) Sep 1996, 142 ( Pt 9)  
p2561-6, ISSN 1350-0872 Journal Code: 9430468

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Subfile: INDEX MEDICUS

The plc gene, which encodes phospholipase C (alpha-toxin) of *Clostridium perfringens*, possesses three poly(A) tracts forming an intrinsically curved DNA region immediately upstream of the promoter. The in vivo transcriptional activity of the plasmid-borne plc gene was stimulated by this curved-DNA-containing sequence, depending on its proper linear and rotational orientation. The in vitro transcriptional activity of the plc gene was also stimulated by the upstream sequence. In addition, the stimulatory effect of the sequence and the degree of DNA bending were greater at lower temperature, as was demonstrated by both in vitro and in vivo transcription assays, and a gel-mobility assay, respectively. A similar temperature effect was also observed with the chromosomal plc gene. These observations suggest that the upstream DNA curvature per se stimulates the initiation of transcription of the plc gene, possibly through direct contact with RNA polymerase.

Tags: Support, Non-U.S. Gov't

Descriptors: \**Clostridium perfringens*--genetics--GE; \*Phospholipase C--genetics--GE; Base Sequence; Chromosome Mapping; Chromosomes--genetics--GE; Chromosomes--physiology--PH; DNA--physiology--PH; Gene Expression Regulation, Bacterial; Molecular Sequence Data; Mutagenesis, Insertional; Mutagenesis, Site-Directed; Nucleic Acid Conformation; Plasmids--genetics--GE; Plasmids--physiology--PH; Promoter Regions (Genetics); Sequence Deletion; Temperature; Transcription, Genetic

CAS Registry No.: 0 (Plasmids); 9007-49-2 (DNA)

Enzyme No.: EC 3.1.4.3 (Phospholipase C)

Record Date Created: 19970113

Record Date Completed: 19970113

10/9/4 (Item 4 from file: 155)

DIALOG(R) File 155:MEDLINE(R)

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09432452 PMID: 1522810

**Role of the upstream region containing an intrinsic DNA curvature in the negative regulation of the phospholipase C gene of *Clostridium perfringens***

Toyonaga T; Matsushita O; Katayama S; Minami J; Okabe A

Department of Microbiology, Kagawa Medical School, Japan.

Microbiology and immunology (JAPAN) 1992, 36 (6) p603-13, ISSN 0385-5600 Journal Code: 7703966

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Subfile: INDEX MEDICUS

The phospholipase C (alpha-toxin) gene (plc) of *Clostridium perfringens* was cloned into pUC19 and the effects of the upstream regions on expression of the plc gene were examined in *Escherichia coli* JM109. When the 0.7-kb region just upstream of the putative -35 site of the gene was deleted, production of phospholipase C increased approximately 10-fold. Northern blot hybridization analysis of the plc transcript showed that the upstream region inhibited **transcription** from the plc promoter. Nucleotide sequencing of this upstream region revealed that there are three periodically repeated (dA)5-6 tracts between positions -66 and -40 of the plc gene. A fragment containing this sequence showed anomalously slow electrophoretic mobility at low temperature, indicating that the region immediately upstream of the plc promoter is a locus of sequence directed DNA-bending. Nested deletions of the upstream region were created from its 5' end by exonuclease III and the effects of deletions on the expression of the plc gene were examined. When the 77-bp fragment containing the two (dA)5-6 tracts were deleted, phospholipase C production increased markedly. These results indicate that the intrinsic DNA curvature upstream of the plc promoter is involved in the negative regulation of the plc gene transcription.

Tags: Comparative Study; Support, Non-U.S. Gov't

Descriptors: \*Bacterial Toxins--genetics--GE; \**Clostridium perfringens* --genetics--GE; \*DNA, Bacterial--genetics--GE; \*Gene Expression Regulation, Enzymologic; \*Genes, Bacterial--genetics--GE; \*Phospholipase C--genetics --GE; Base Sequence; Blotting, Northern; Chromosome Deletion; *Clostridium perfringens*--enzymology--EN; *Escherichia coli*--genetics--GE; Molecular Sequence Data; Phospholipase C--metabolism--ME; Plasmids--genetics--GE; RNA, Messenger--metabolism--ME; Restriction Mapping; Transfection

CAS Registry No.: 0 (Bacterial Toxins); 0 (DNA, Bacterial); 0 (Plasmids); 0 (RNA, Messenger)

Enzyme No.: EC 3.1.4.- (*Clostridium perfringens* alpha-toxin); EC 3.1.4.3 (Phospholipase C)

Record Date Created: 19921015

Record Date Completed: 19921015

10/9/3 (Item 3 from file: 144)

DIALOG(R) File 144:Pascal

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09115301 PASCAL No.: 90-0283682

**Gene cloning shows the alpha-toxin of *Clostridium perfringens* to contain both sphingomyelinase and lecithinase activities**

SAINT-JOANIS B; GARNIER T; COLE S T

Inst. Pasteur, Paris 75724, France

Journal: MGG. Molecular & general Genetics, 1989, 219 (3) 453-460

ISSN: 0026-8925 CODEN: MGGEAE Availability: CNRS-3571

No. of Refs.: 2 p.

Document Type: P (Serial) ; A (Analytic)

Country of Publication: Federal Republic of Germany

Language: English

The plc gene encoding the alpha-toxin of *Clostridium perfringens*, has been cloned, sequenced and expressed in *Escherichia coli*. Transcriptional analysis of mRNAs produced in vivo by *C. perfringens* and *E. coli*, and in vitro using purified RNA polymerase from *C. perfringens* revealed that plc is transcribed constitutively from a single promoter. Enzymological studies with the amplified plc gene product unambiguously demonstrated that both lecithinase (phospholipase C) and sphingomyelinase activities were associated with this 43000 dalton cytotoxin

English Descriptors: Nucleotide sequence; Gene expression; Molecular cloning; DNA; Transcription; Transcription promoter; Phospholipase C; Sphingomyelin phosphodiesterase; Comparative study; Genetic transformation; In vitro transcription; Radiolabelling; Gel electrophoresis; Enzymatic activity; *Clostridium perfringens*; Toxin; Enzyme; Primer extension technique

Broad Descriptors: Clostridiaceae; Clostridiales; Bacteria; Clostridiaceae; Clostridiales; Bacterie; Clostridiaceae; Clostridiales; Bacteria

French Descriptors: Sequence nucleotide; Expression genique; Clonage  
moleculaire; DNA; Transcription; Promoteur transcription; Phospholipase C  
; Sphingomyelin phosphodiesterase; Etude comparative; Transformation  
genetique; Transcription in vitro; Marquage radioisotopique;  
Electrophorese gel; Activite enzymatique; Clostridium perfringens; Toxine  
; Enzyme; Toxine alpha ; Gene plc; Technique extension amorce

Classification Codes: 002A04C02

10/9/5 (Item 5 from file: 34)

DIALOG(R)File 34:SciSearch(R) Cited Ref Sci

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03084944 Genuine Article#: NB994 Number of References: 32

**Title: THE VIRR GENE, A MEMBER OF A CLASS OF 2-COMPONENT RESPONSE  
REGULATORS, REGULATES THE PRODUCTION OF PERFRINGOLYSIN -O,  
COLLAGENASE, AND HEMAGGLUTININ IN CLOSTRIDIUM - PERFRINGENS**

Author(s): SHIMIZU T; BATHEIN W; TAMAKI M; HAYASHI H

Corporate Source: UNIV TSUKUBA, INST BASIC MED SCI, DEPT MICROBIOL, 1-1-1  
TENOHDAI/TSUKUBA/IBARAKI 305/JAPAN/

Journal: JOURNAL OF BACTERIOLOGY, 1994, V176, N6 (MAR), P1616-1623

ISSN: 0021-9193

Language: ENGLISH Document Type: ARTICLE

Geographic Location: JAPAN

Subfile: SciSearch; CC LIFE--Current Contents, Life Sciences

Journal Subject Category: MICROBIOLOGY

**Abstract:** The perfringolysin O (theta-toxin) gene (pfoA) of Clostridium  
perfringens was cloned into an Escherichia coli-C. perfringens shuttle  
vector, and the pfoA gene was expressed in mutants of C. perfringens 13  
which lacked the production of perfringolysin O. One group (SI117)  
could express the pfoA gene, and tile other (SI112) could not. A  
mutation in the regulatory system for pfoA gene expression was  
suspected in SI112. A chromosomal DNA library constructed from strain  
13 was transformed into strain SI112 to identify the regulatory gene(s)  
for the pfoA gene. Five strains of 10,000 transformants restored  
perfringolysin O production. All contained a 2.5-kb DNA fragment.  
This fragment activated the transcription of the pfoA gene and also  
restored the production of collagenase (kappa-toxin) and hemagglutinin  
in strain SI112. Deletion analysis showed that a 1.25-kb region was  
sufficient for the trans activity, and sequence analysis disclosed that  
open reading frame 2 (ORF2) was located in this region. A homology  
search for the deduced amino acid sequence revealed that ORF2 was  
homologous to a response regulator in a two-component signal  
transduction system. ORF2 was designated virR, and it is suggested that  
the virR gene plays an important role in the pathogenicity of C.  
**perfringens .**

Identifiers--KeyWords Plus: ELECTROPORATION-INDUCED TRANSFORMATION;  
PHOSPHOLIPASE-C GENE; NUCLEOTIDE -SEQUENCE; PLASMID; CLONING;  
EXPRESSION; FRAGMENTS; CELLS

Research Fronts: 92-4812 003 (PUTATIVE ANAEROBIC COPROPORPHYRINOGEN-III  
OXIDASE IN RHODOBACTER-SPHAEROIDES; TRANSCRIPTIONAL REGULATORY ELEMENT;  
FUNCTIONAL EXPRESSION)

92-2989 001 (PHOSPHORYLATION OF BACTERIAL RESPONSE REGULATOR PROTEINS;  
FLAGELLAR SWITCH MUTATIONS; BACILLUS-SUBTILIS CHEMOTAXIS; INVITRO  
TRANSCRIPTION ; PROMOTER REGION)

92-3896 001 (EXTRACELLULAR COLLAGENOLYTIC PROTEINASES; SERUM  
COLLAGENASE; PORPHYROMONAS-GINGIVALIS PRTC GENE)

92-8079 001 (ESCHERICHIA-COLI GENE; CHARACTERIZATION OF DNA  
DUMBBELLS; EXPRESSION SIGNALS)

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10/9/6 (Item 6 from file: 35)

DIALOG(R)File 35:Dissertation Abs Online

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01601066 ORDER NO: NOT AVAILABLE FROM UNIVERSITY MICROFILMS INT'L.

**DEVELOPMENT OF A NOVEL EXPRESSION SYSTEM IN CLOSTRIDIUM PERFRINGENS  
(GENE EXPRESSION, SHUTTLE VECTOR)**

Author: BROWN, ROBERT CHRISTOPHER

Degree: PH.D.

Year: 1997

Corporate Source/Institution: OPEN UNIVERSITY (UNITED KINGDOM) (0949)

Source: VOLUME 58/04-C OF DISSERTATION ABSTRACTS INTERNATIONAL.

PAGE 1203.

Descriptors: BIOLOGY, MOLECULAR ; BIOLOGY, MICROBIOLOGY

Descriptor Codes: 0307; 0410

Genetic manipulation and recombinant expression in the genus *Clostridium* are still in their infancy when compared to the technology developed for *Escherichia coli*. As the biotechnological importance of clostridia for commercial and pharmaceutical exploitation has become apparent, research has intensified. *C. perfringens* is a member of this genus, as are the neurotoxin producer *C. botulinum*, the opportunistic pathogen *C. difficile*, and the solventogenic *C. acetobutylicum*. *C. perfringens* is of great clinical importance as the causative agent in many human and animal diseases. These diseases are mediated by extracellular enzymes or toxins. The study of these toxins and their regulation has accelerated genetic transfer techniques in this pathogenic organism, as well as elucidating some of the mechanisms of pathogenesis.

A range of shuttle-vectors has been developed for *C. perfringens*. The potential to secrete recombinant proteins, combined with the relatively short doubling time (ca. 20 minutes), make it a suitable candidate for Gram-positive recombinant DNA technology.

The production of recombinant non-toxic fragments of *C. botulinum* neurotoxin type A (BoNT/A) for vaccine and therapeutic development has been of high priority within this laboratory for a number of years. The expression of recombinant BoNT/A has proven problematic in the recombinant host *E. coli*, due to cytotoxic effects, codon usage and proteolytic

activity. The optimum host for the production of recombinant BoNT/A fragments would be *C. botulinum*. However, because of safety considerations, and primarily due to the lack of an established gene transfer technique in this organism, this avenue has not yet been pursued. An alternative recombinant clostridial host may prove a way of circumventing problems of gene transfer, while attaining a high degree of authentic recombinant product. *C. perfringens* was examined as the alternative clostridial recombinant host.

A range of established shuttle-vectors for *C. perfringens* were examined, as well as vectors developed in other Gram-positive bacteria. This investigation served as a basis for the optimisation of electrotransformation of *C. perfringens*, and determined the stability and potential of utilising these vectors within a recombinant expression system.

Problems of vector instability, both structural and segregational lead to the development of a recombination system to integrate an expression cassette within the *C. perfringens* genome. The target for integration was the *recA* gene, the recombination locus that would integrate via flanking *recA* homologues of the expression cassette. Initially reporter gene fragments were recombined with the *C. perfringens* genome as an indication of integration, by the exhibition of chloramphenicol resistance and elevated Lac<sup>+</sup> phenotype. Finally, **clostridial promoter** elements for **transcription** and translation were incorporated within the expression cassette to control the production of recombinant fragments of BoNT/A. A secretory leader sequence for export of recombinant protein was an additional component of this expression cassette. Recombinant fusion proteins comprising non-toxin BoNT/A fragments associated with N-terminal peptides to facilitate purification were successfully expressed in *C. perfringens* strain 13. This procedure marks the first demonstration of **heterologous DNA** expression in *C. perfringens* and the production of recombinant **clostridial** non-toxic BoNT/A fragments.

10/9/7 (Item 7 from file: 144)  
DIALOG(R) File 144:Pascal  
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08602368 PASCAL No.: 89-0151446

**Identification and molecular genetic analysis of replication functions of the bacteriocinogenic plasmid pIP404 from *Clostridium perfringens***

GARNIER T; COLE S T

Inst. Pasteur, Paris 75724, France

Journal: Plasmid, 1988, 19 (2) 151-160

ISSN: 0147-619X CODEN: PLSMDX Availability: CNRS-17779

No. of Refs.: 2 p.

Document Type: P (Serial) ; A (Analytic)

Country of Publication: USA

Language: English



English Descriptors: *Clostridium perfringens*; Replication; Molecular cloning; Gene; Antisense RNA; Transcription promoter; Bacteriocinogeny; Origin; Repeated sequence

Broad Descriptors: Clostridiaceae; Clostridiales; Bacteria; Clostridiaceae; Clostridiales; Bacterie; Clostridiaceae; **Clostridiales** ; Bacteria

French Descriptors: *Clostridium perfringens*; Replication; Clonage moleculaire; Gene; RNA antisens; Promoteur transcription; Bacteriocinogenie; Origine; Sequence repetee; Plasmide pIP404; Proteine cop; Proteine rcp

Classification Codes: 002A05B09; 215C02A03

10/9/8 (Item 8 from file: 5)  
DIALOG(R) File 5:Biosis Previews(R)  
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0006699658 BIOSIS NO.: 198988014773

**NUCLEOTIDE SEQUENCE ANALYSIS AND EXPRESSION STUDIES OF A CHLORAMPHENICOL ACETYLTRANSFERASE-CODING GENE FROM CLOSTRIDIUM-PERFRINGENS**

AUTHOR: STEFFEN C (Reprint); MATZURA H

AUTHOR ADDRESS: MOLEKULARE GENETIK IM NEUENHEIMER FELD 230, D-6900

HEIDELBERG, WEST GERMANY\*\*WEST GERMANY

JOURNAL: Gene (Amsterdam) 75 (2): p349-354 1989

ISSN: 0378-1119

DOCUMENT TYPE: Article

RECORD TYPE: Abstract

LANGUAGE: ENGLISH

**ABSTRACT:** The nucleotide sequence of a CmR determinant, located on the **Clostridium perfringens** plasmid pIP401, was determined and its gene product was identified as chloramphenicol-acetyltransferase (CAT). The cat structural gene is preceded by transcription-initiation signals characteristic for *Escherichia coli* .sigma.70 or *Bacillus subtilis* .sigma.43 promoters. By promoter probing in the **heterologous** hosts the direction of transcription of the **clostridial** cat gene was analysed and the cat mRNA start point was determined in vitro using the RNA polymerases of *E. coli* and *B. subtilis*. Comparison of the amino acid sequences of *C. perfringens* CAT and other CAT proteins of Gram-positive and Gram-negative origin shows a remarkable degree of homology between the various enzymes.

REGISTRY NUMBERS: 9040-07-7: CHLORAMPHENICOL ACETYLTRANSFERASE

DESCRIPTORS: BACILLUS-SUBTILIS ESCHERICHIA-COLI **TRANSCRIPTION** INITIATION

**PROMOTER** MOLECULAR SEQUENCE DATA DEDUCED AMINO ACID SEQUENCE

DESCRIPTORS:

MAJOR CONCEPTS: Biochemistry and Molecular Biophysics; Enzymology--

Biochemistry and Molecular Biophysics; Genetics; Metabolism; Molecular

Genetics--Biochemistry and Molecular Biophysics; Physiology

BIOSYSTEMATIC NAMES: Enterobacteriaceae--Facultatively Anaerobic

Gram-Negative Rods, Eubacteria, Bacteria, Microorganisms;

Endospore-forming Gram-Positives--Eubacteria, Bacteria, Microorganisms

COMMON TAXONOMIC TERMS: Bacteria; Eubacteria; Microorganisms

CHEMICALS & BIOCHEMICALS: CHLORAMPHENICOL ACETYLTRANSFERASE

CONCEPT CODES:

10010 Comparative biochemistry

10062 Biochemistry studies - Nucleic acids, purines and pyrimidines

10064 Biochemistry studies - Proteins, peptides and amino acids

10300 Replication, transcription, translation

10506 Biophysics - Molecular properties and macromolecules

10802 Enzymes - General and comparative studies: coenzymes

10806 Enzymes - Chemical and physical

10808 Enzymes - Physiological studies

13014 Metabolism - Nucleic acids, purines and pyrimidines

22002 Pharmacology - General

31000 Physiology and biochemistry of bacteria

31500 Genetics of bacteria and viruses

38504 Chemotherapy - Antibacterial agents

BIOSYSTEMATIC CODES:

06702 Enterobacteriaceae

07810 Endospore-forming Gram-Positives

10/9/9 (Item 9 from file: 155)

DIALOG(R) File 155:MEDLINE(R)

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12832817 PMID: 8566714

**Transcriptional analysis of the beta-galactosidase gene (pbg) in Clostridium perfringens.**

Kobayashi T; Shimizu T; Hayashi H

Department of Microbiology, University of Tsukuba, Ibaraki, Japan.

FEMS microbiology letters (NETHERLANDS) Nov 1 1995, 133 (1-2) p65-9,

ISSN 0378-1097 Journal Code: 7705721

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Subfile: INDEX MEDICUS

The mode of expression of the beta-galactosidase gene (pbg) of *Clostridium perfringens* was examined. The pbg gene was transcribed on a single 3.7-kb mRNA. The transcript contained a message for ORF54, located upstream of the pbg gene in the chromosome, indicating that ORF54 and the pbg gene comprise one operon (pbg operon). Expression of the pbg operon was induced by lactose at the transcriptional level. The promoter structure of the pbg operon was characterized by many palindrome structures and direct repeats, which suggests that there might be some catabolite regulation of the expression of the pbg operon in *C. perfringens*.

Tags: Support, Non-U.S. Gov't

Descriptors: *Clostridium perfringens* --genetics--GE; \*Genes, Bacterial --genetics--GE; \*Transcription, Genetic--genetics--GE; \*beta-Galactosidase --genetics--GE; Base Sequence; Chromosome Mapping; Cloning, Molecular; *Clostridium perfringens*--enzymology--EN; Lactose--metabolism--ME; Molecular Sequence Data; RNA, Bacterial--analysis--AN; RNA, Messenger--analysis--AN

CAS Registry No.: 0 (RNA, Bacterial); 0 (RNA, Messenger); 63-42-3 (Lactose)

Enzyme No.: EC 3.2.1.23 (beta-Galactosidase)

Record Date Created: 19960301

Record Date Completed: 19960301

10/9/10 (Item 10 from file: 144)

DIALOG(R) File 144:Pascal

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09302147 PASCAL No.: 91-0092521

**Cloning and sequencing of the genes encoding acid-soluble spore proteins from *Clostridium perfringens***

HOLCK A; BLOM H; GRANUM P E

Norwegian food res. inst., As 1430, Norway

Journal: Gene, 1990, 91 (1) 107-111

ISSN: 0378-1119 CODEN: GENED6 Availability: INIST-17570;  
354000009725550150/NUM

No. of Refs.: 15 ref.

Document Type: P (Serial) ; A (Analytic)

Country of Publication: Netherlands

Language: English

English Descriptors: Spores; Proteins; Nucleotide sequence; DNA; Molecular cloning; Homology; Transcription promoter; *Clostridium perfringens*

Broad Descriptors: **Clostridiaceae ; Clostridiales ; Bacteria;**

**Clostridiaceae ; Clostridiales ; Bacterie; Clostridiaceae ;**

**Clostridiales ; Bacteria**

French Descriptors: Spore; Proteine; Sequence nucleotide; DNA; Clonage moléculaire; Homologie; Promoteur transcription; *Clostridium perfringens*; Proteine ASSP; Gene sspC1; Gene sspC2

Classification Codes: 002A04C02

10/9/12 (Item 12 from file: 34)

DIALOG(R) File 34:SciSearch(R) Cited Ref Sci

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03238985 Genuine Article#: NP484 Number of References: 80

**Title: IDENTIFICATION AND MOLECULAR ANALYSIS OF A LOCUS THAT REGULATES EXTRACELLULAR TOXIN PRODUCTION IN CLOSTRIDIUM - PERFRINGENS**

Author(s): LYRISTIS M; BRYANT AE; SLOAN J; AWAD MM; NISBET IT; STEVENS DL;

ROOD JI

Corporate Source: MONASH UNIV, DEPT MICROBIOL/CLAYTON/VIC 3168/AUSTRALIA/;  
MONASH UNIV, DEPT MICROBIOL/CLAYTON/VIC 3168/AUSTRALIA/; VET ADM MED  
CTR, INFECT DIS RES UNIT/BOISE//ID/83702; UNIV WASHINGTON, SCH MED, DEPT  
MED/SEATTLE//WA/98195; COMMONWEALTH SERUM LABS/PARKVILLE/VIC  
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Journal: MOLECULAR MICROBIOLOGY, 1994, V12, N5 (JUN), P761-777

ISSN: 0950-382X

Language: ENGLISH Document Type: ARTICLE

Geographic Location: USA; AUSTRALIA

Subfile: SciSearch; CC LIFE--Current Contents, Life Sciences

Journal Subject Category: BIOCHEMISTRY & MOLECULAR BIOLOGY; MICROBIOLOGY

Abstract: The anaerobic bacterium *Clostridium perfringens* mediates clostridial myonecrosis, or gas gangrene, by producing a number of extracellular toxins and enzymes. Transposon mutagenesis with Tn916 was used to isolate a pleiotropic mutant of *C. perfringens* that produced reduced levels of phospholipase C, protease and sialidase, and did not produce any detectable perfringolysin O activity. Southern hybridization revealed that a single copy of Tn916 had inserted into a 2.7 kb HindIII fragment in the *C. perfringens* chromosome. A 4.3kb PstI fragment, which spanned the Tn916 insertion site, was cloned from the wild-type strain. When subcloned into a shuttle vector and introduced into *C. perfringens* this fragment was able to complement the Tn916-derived mutation. Transformation of the mutant with plasmids containing the 2.7kb HindIII fragment, or the 4.3kb PstI fragment, resulted in toxin and enzyme levels greater than or equal to those of the wild-type strain. The PstI fragment was sequenced and found to potentially encode seven open reading frames, two of which appeared to be arranged in an operon and shared sequence similarity with members of two-component signal transduction systems. The putative virR gene encoded a protein with a deduced molecular weight of 30140, and with sequence similarity to activators in the response regulator family of proteins. The next gene, virS, into which Tn916 had inserted, was predicted to encode a membrane-spanning protein with a deduced molecular weight of 51274. The putative VirS protein had sequence similarity to sensor proteins and also contained a histidine residue highly conserved in the histidine protein kinase family of sensor proteins. Virulence studies carried out using a mouse model implicated the virS gene in the pathogenesis of histotoxic *C. perfringens* infections. It was concluded that a two-component sensor regulator system that activated the expression of a number of extracellular toxins and enzymes involved in virulence had been cloned and sequenced. A model that described the regulation of extracellular toxin production in *C. perfringens* was constructed.

Identifiers--KeyWords Plus: PHOSPHOLIPASE-C GENE; TRANSFERABLE TETRACYCLINE RESISTANCE; EXPERIMENTAL GAS-GANGRENE; O THETA-TOXIN; ESCHERICHIA-COLI; NUCLEOTIDE -SEQUENCE; ALPHA-TOXIN; AGROBACTERIUM-TUMEFACIENS; SIGNAL TRANSDUCTION; PHOSPHATE REGULON

Research Fronts: 92-2989 003 (PHOSPHORYLATION OF BACTERIAL RESPONSE REGULATOR PROTEINS; FLAGELLAR SWITCH MUTATIONS; BACILLUS-SUBTILIS CHEMOTAXIS; INVITRO TRANSCRIPTION ; PROMOTER REGION)

92-3160 001 (HYBRIDIZATION OF DNA ; PROMOTER REGION; MOLECULAR CLONES; EFFICIENT INITIATION; INTERACTIVE SYSTEM; STRUCTURAL ELEMENTS; RIBOSOMAL-PROTEIN OPERON)

92-4812 001 (PUTATIVE ANAEROBIC COPROPORPHYRINOGEN-III OXIDASE IN RHODOBACTER-SPHAEROIDES; TRANSCRIPTIONAL REGULATORY ELEMENT; FUNCTIONAL EXPRESSION)

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DIALOG(R)File 34:SciSearch(R) Cited Ref Sci  
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01195255 Genuine Article#: GD031 Number of References: 29

Title: **CLONING, MAPPING, AND MOLECULAR CHARACTERIZATION OF THE RIBOSOMAL-  
RNA OPERONS OF CLOSTRIDIUM - PERFRINGENS**

Author(s): GARNIER T; CANARD B; COLE ST

Corporate Source: INST PASTEUR, GENET MOLEC BACTERIENNE LAB, 28 RUE DOCTEUR  
ROUX/F-75724 PARIS 15//FRANCE//; INST PASTEUR, GENET MOLEC BACTERIENNE  
LAB, 28 RUE DOCTEUR ROUX/F-75724 PARIS 15//FRANCE/

Journal: JOURNAL OF BACTERIOLOGY, (1991) V173, N17, P5431-5438

Language: ENGLISH Document Type: ARTICLE

Geographic Location: FRANCE

Subfile: SciSearch; CC LIFE--Current Contents, Life Sciences

Journal Subject Category: MICROBIOLOGY

Abstract: All 10 rRNA operons have been situated on the genome map of the  
anaerobic pathogen Clostridium perfringens. Four of these have been  
cloned and partially sequenced, and their transcriptional patterns in  
vivo and in vitro have been examined. Expression of rrnA, rrnB, and  
rrnE is directed by tandem promoters, P1 and P2, whereas rrnH is the  
only one to be expressed from a single promoter, which resembles P1. On  
inspection of the nucleotide sequences of the control regions, several  
sites which might be involved in the regulation of rrn expression were  
identified. These include a possible upstream activating region which  
could be recognized by the C. perfringens equivalent of the Escherichia  
coli Fis protein and a stringent response target site. Studies of  
maturation of 16S RNA identified two 5' cleavage sites and sequence  
analysis showed the dG+dC content of its gene, rrs, to be 52%, which is  
twice that of the genome.

Identifiers--KeyWords Plus: RIBOSOMAL- RNA OPERONS; BACILLUS-SUBTILIS;  
DNA -SEQUENCE; TRANSCRIPTION; ORGANIZATION; PROMOTER; REGION; 16S;  
VECTORS; INVITRO

Research Fronts: 89-1447 002 (DEVELOPMENTALLY REGULATED GENE; CAPPING  
PROTEIN; CDNA SEQUENCE; GENOME ORGANIZATION)

89-0639 001 ( GENETIC DIVERSITY; COMPLEX POPULATION DIFFERENTIATION;  
ATLANTIC SALMON; EASTERN NORTH-AMERICA; ENZYME ELECTROPHORESIS)

89-1508 001 (ESCHERICHIA-COLI CHROMOSOME; DNAK GENE; PUREK OPERON  
ENCODING 5'-PHOSPHORIBOSYL-5-AMINOIMIDAZOLE CARBOXYLASE)

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DIALOG(R) File 34:SciSearch(R) Cited Ref Sci  
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00847254 Genuine Article#: FA766 Number of References: 40

Title: **RELATIONSHIP BETWEEN THE CLOSTRIDIUM - PERFRINGENS CATQ**

**GENE-PRODUCT AND CHLORAMPHENICOL ACETYLTRANSFERASES FROM OTHER BACTERIA**

Author(s): BANNAM TL; ROOD JI

Corporate Source: MONASH UNIV, DEPT MICROBIOL/CLAYTON/VIC 3168/AUSTRALIA/;

MONASH UNIV, DEPT MICROBIOL/CLAYTON/VIC 3168/AUSTRALIA/

Journal: ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, 1991, V35, N3, P471-476

Language: ENGLISH Document Type: ARTICLE

Geographic Location: AUSTRALIA

Subfile: SciSearch; CC LIFE--Current Contents, Life Sciences

Journal Subject Category: MICROBIOLOGY; PHARMACOLOGY & PHARMACY

Abstract: The nucleotide sequence of the *Clostridium perfringens*

chloramphenicol acetyltransferase (CAT)-encoding resistance determinant, *catQ*, was determined. An open reading frame encoding a protein of 219 amino acids with a molecular weight of 26,014 was identified. Although *catQ* was expressed constitutively, sequences similar in structure to those found upstream of inducible *cat* genes were observed. The *catQ* gene was distinct from the *C. perfringens* *catP* determinant. The deduced CATQ monomer had considerable amino acid sequence conservation compared with CATP (53% similarity) and other known CAT protein (39 to 53%). Phylogenetic analysis revealed that the CATQ monomer was as closely related to CAT proteins from *Staphylococcus aureus* and *Campylobacter coli* as it was to CAT monomer from the **clostridia**.

Identifiers--KeyWords Plus: **NUCLEOTIDE** -SEQUENCE ANALYSIS; HYBRIDIZATION ANALYSIS; ACETYL TRANSFERASE; BACILLUS-SUBTILIS; ESCHERICHIA-COLI; ACTIVE-SITE; RESISTANCE; EXPRESSION; **PLASMIDS**; CLONING

Research Fronts: 89-1447 001 (DEVELOPMENTALLY REGULATED GENE; CAPPING PROTEIN; **CDNA** SEQUENCE; GENOME ORGANIZATION)

89-3723 001 (ESCHERICHIA-COLI K-12; MALTOSE-BINDING PROTEIN; OSMOTIC REGULATION OF PORIN EXPRESSION)

89-6034 001 (BACILLUS-SUBTILIS CHROMOSOME; REPLICATION ORIGINS OF SINGLE-STRANDED- **DNA** **PLASMID** PUB110; LACTOCOCCUS-LACTIS GENE; PROTOPLAST TRANSFORMATION)

89-6184 001 (ESCHERICHIA-COLI **PROMOTERS** ; REGULATION OF **TRANSCRIPTION** ; ERWINIA-CHRYSANTHEMI GENE ENCODING 2-KETO-3-DEOXYGLUCONATE PERMEASE)

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DIALOG(R) File 434:SciSearch(R) Cited Ref Sci  
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09269425 Genuine Article#: R9221 Number of References: 50

**Title: MOLECULAR-CLONING AND NUCLEOTIDE -SEQUENCE OF THE ALPHA-TOXIN  
 (PHOSPHOLIPASE-C) OF CLOSTRIDIUM - PERFRINGENS**

**Author(s): TITBALL RW; HUNTER SEC; MARTIN KL; MORRIS BC; SHUTTLEWORTH AD;  
 RUBIDGE T; ANDERSON DW; KELLY DC**

**Corporate Source: CHEM DEF ESTAB/SALISBURY SP4 OJQ/WILTS/ENGLAND/**

**Journal: INFECTION AND IMMUNITY, 1989, V57, N2, P367-376**

**Language: ENGLISH Document Type: ARTICLE**

**Geographic Location: ENGLAND**

**Subfile: SciSearch; CC LIFE--Current Contents, Life Sciences**

**Journal Subject Category: IMMUNOLOGY**

**Research Fronts: 87-3538 003 (ESCHERICHIA-COLI RNA -POLYMERASE;**

**PROMOTER RECOGNITION; STRUCTURAL GENE; TRANSCRIPTION INITIATION;  
 NUCLEOTIDE -SEQUENCE HOMOLOGIES; TRANSLATIONAL REQUIREMENT)**

**87-3127 002 (CALCIUM-BINDING PROTEIN; TISSUE LOCALIZATION;**

**ESCHERICHIA-COLI GENE; PROGESTERONE-RECEPTOR REGULATION)**

**87-0968 001 (CONFORMATION OF SHORT LINEAR PEPTIDES; IONIC SOLVATION IN  
 WATER COSOLVENT MIXTURES; PROTEIN FOLDING; REFINED CRYSTAL-STRUCTURE;  
 HYDROPHOBIC INTERACTIONS)**

**87-1403 001 (OPIN GENE; MALIC ENZYME MESSENGER- RNA ; CDNA CLONES;  
 STRUCTURAL ORGANIZATION; DISTINCT FORMS)**

**87-3213 001 (STRICTLY ANAEROBIC BACTERIUM; DEOXYRIBONUCLEIC-ACID  
 HYBRIDIZATION; NUMERICAL TAXONOMY; ESCHERICHIA-COLI GENE)**

**87-8007 001 (OUTER-MEMBRANE OF ESCHERICHIA-COLI; PROTEIN ANTIGEN;  
 LEADER PEPTIDASE)**

**87-8061 001 (SECRETION SIGNAL SEQUENCE; HERPES-SIMPLEX VIRUS TYPE-1;  
 MALTOS-BINDING PROTEIN; SHIGA-LIKE TOXIN GENES OF ESCHERICHIA-COLI;  
 MESSENGER- RNA EXPRESSION)**

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**CLONING MAPPING AND MOLECULAR CHARACTERIZATION OF THE RNA OPERONS OF  
CLOSTRIDIUM -PERFRINGENS**

AUTHOR: GARNIER T (Reprint); CANARD B; COLE S T

AUTHOR ADDRESS: LABORATOIRE DE GENETIQUE MOLECULAIRE BACTERIENNE, INSTITUT

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JOURNAL: Journal of Bacteriology 173 (17): p5431-5438 1991

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RECORD TYPE: Abstract

LANGUAGE: ENGLISH

**ABSTRACT:** All 10 rRNA operons have been situated on the genome map of the anaerobic pathogen *Clostridium perfringens*. Four of these have been cloned and partially sequenced, and their transcriptional patterns in vivo and in vitro have been examined. Expression of *rrnA*, *rrnB*, and *rrnE* is directed by tandem promoters, P1 and P2, whereas *rrnH* is the only one to be expressed from a single promoter, which resembles P1. On inspection of the nucleotide sequences of the control regions, several sites which

might be involved in the regulation of *rrn* expression were identified. These include a possible upstream activating region which could be recognized by the *C. perfringens* equivalent of the *Escherichia coli* Fis protein and a stringent response target site. Studies of maturation of 16S RNA identified two 5' cleavage sites and sequence analysis showed the dG+dC content of its gene, *rrs*, to be 52%, which is twice that of the genome.

REGISTRY NUMBERS: 140083-05-2: M69264; 139850-63-8: M69267  
DESCRIPTORS: ESCHERICHIA-COLI FIS HOMOLOG TRANSCRIPTION REGULATORY REGIONS  
PROMOTER USE BASE COMPOSITION GENBANK-M69265 GENBANK-M69266 GENBANK-M69264  
GENBANK-M69267 NUCLEOTIDE SEQUENCE MOLECULAR SEQUENCE DATA  
DESCRIPTORS:

MAJOR CONCEPTS: Biochemistry and Molecular Biophysics; Genetics;  
Metabolism; Molecular Genetics--Biochemistry and Molecular Biophysics  
BIOSYSTEMATIC NAMES: Enterobacteriaceae--Facultatively Anaerobic  
Gram-Negative Rods, Eubacteria, Bacteria, Microorganisms;  
Endospore-forming Gram-Positives--Eubacteria, Bacteria, Microorganisms  
COMMON TAXONOMIC TERMS: Bacteria; Eubacteria; Microorganisms

MOLECULAR SEQUENCE DATABANK NUMBER: M69264--GENBANK; M69267--GENBANK

CONCEPT CODES:

10062 Biochemistry studies - Nucleic acids, purines and pyrimidines  
10064 Biochemistry studies - Proteins, peptides and amino acids  
10300 Replication, transcription, translation  
10506 Biophysics - Molecular properties and macromolecules  
13014 Metabolism - Nucleic acids, purines and pyrimidines  
31000 Physiology and biochemistry of bacteria  
31500 Genetics of bacteria and viruses

BIOSYSTEMATIC CODES:

06702 Enterobacteriaceae  
07810 Endospore-forming Gram-Positives

10/9/18 (Item 18 from file: 144)

DIALOG(R) File 144:Pascal

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08706627 PASCAL No.: 89-0255883

**Studies of UV-inducible promoters from *Clostridium perfringens* in vivo and in vitro**

GARNIER T; COLE S T

Inst. Pasteur, Paris 75724, France

Journal: Molecular microbiology, 1988, 2 (5) 607-614

ISSN: 0950-382X Availability: CNRS-21344

No. of Refs.: 34 ref.

Document Type: P (Serial) ; A (Analytic)

Country of Publication: United Kingdom

Language: English

English Descriptors: Gene expression; Gene; Bacteriocin; Transcription promoter; Ultraviolet irradiation; Induction; Nucleotide sequence; *Clostridium perfringens*

Broad Descriptors: **Clostridiaceae ; Clostridiales ; Bacteria; Clostridiaceae ; Clostridiales ; Bacterie; Clostridiaceae ; Clostridiales ; Bacteria**

French Descriptors: Expression genique; Gene; Bacteriocine; Promoteur transcription; Irradiation UV; Induction; Sequence nucleotide; *Clostridium perfringens*

Classification Codes: 002A04C02

10/9/20 (Item 20 from file: 34)

DIALOG(R) File 34:SciSearch(R) Cited Ref Sci

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03302660 Genuine Article#: NU575 Number of References: 51

**Title: ORGANIZATION OF THE BOTULINUM NEUROTOXIN C1 GENE AND ITS ASSOCIATED  
NONTOXIC PROTEIN GENES IN CLOSTRIDIUM-BOTULINUM-C-468**

Author(s): HAUSER D; EKLUND MW; BOQUET P; POPOFF MR

Corporate Source: INST PASTEUR,UNITE ANAEROBIES,25 RUE DOCTEUR ROUX/F-75724  
PARIS 15//FRANCE/; INST PASTEUR,UNITE TOXINES MICROBIENNES/F-75724PARIS  
15//FRANCE/; NW FISHERIES CTR,DIV UTILIZAT RES/SEATTLE//WA/98112

Journal: MOLECULAR & GENERAL GENETICS, 1994, V243, N6 (JUN 15), P631-640

ISSN: 0026-8925

Language: ENGLISH Document Type: ARTICLE

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Subfile: SciSearch; CC LIFE--Current Contents, Life Sciences

Journal Subject Category: GENETICS & HEREDITY; BIOCHEMISTRY & MOLECULAR  
BIOLOGY

**Abstract:** A 12.3 kb DNA fragment encompassing the botulinum neurotoxin C1 (BoNT/C1) gene and an upstream flanking region was sequenced from *Clostridium botulinum* C 468 phage 1C. The resulting bont/C1 locus includes six genes which are organized into three transcriptional units. Cluster 1 encompasses the bont/C1 gene and an upstream gene encoding a non-toxic protein associated with the toxin (Antp139/C1). Transcriptional analysis revealed that these two genes form an operon; the bont/C1 gene can be transcribed alone or co-transcribed with antp139/C1. Cluster 2 encompasses three genes (antp33/C1, antp17/C1 and antp70/C1), which also form an operon. The corresponding proteins are similar to components of the hemagglutinin complex associated with BoNT/A and BoNT/B of *C. botulinum* A and B. In addition, Antp33/C1 is identical to HA-33, an hemagglutinin encoded by *C. botulinum* C-Stockholm phage C-St; Antp70/C1 displays some relatedness to *C. perfringens* enterotoxin. The third transcriptional unit consists of orf-22, which encodes a basic protein showing 29% identity with the gene product of uviA, a plasmid-encoded protein of 22 kDa which has been identified as a positive regulator of the bacteriocin production in *C. perfringens*. Orf-22 could be an effector controlling the expression of the bont/C1 and its antp genes in *C. botulinum* C 468.

**Descriptors--Author Keywords:** REVERSE TRANSCRIPTASE-POLYMERASE CHAIN  
REACTION (RT-PCR) ; CLOSTRIDIUM BOTULINUM ; BOTULINUM NEUROTOXIN  
COMPLEX

**Identifiers--KeyWords Plus:** COMPLETE NUCLEOTIDE-SEQUENCE;  
AMINO-ACID-SEQUENCE; NEUROTRANSMITTER RELEASE; TETANUS TOXIN;  
PROGENITOR TOXIN; ENCODING GENE; F NEUROTOXIN; BACTERIOPHAGE;  
COMPONENT; STRAINS

**Research Fronts:** 92-4812 002 (PUTATIVE ANAEROBIC COPROPORPHYRINOGEN-III  
OXIDASE IN RHODOBACTER-SPHAEROIDES; TRANSCRIPTIONAL REGULATORY ELEMENT;  
FUNCTIONAL EXPRESSION)

92-2113 001 (DNA CLEAVAGE; ACTIVE-SITE TYROSINE; RAPID DEPROTECTION OF  
SYNTHETIC OLIGONUCLEOTIDES)

92-3447 001 (ESCHERICHIA-COLI RNA-POLYMERASE; INVITRO **TRANSCRIPTION** ;  
PROMOTER MELTING INVIVO)

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10/9/21 (Item 21 from file: 155)

DIALOG(R) File 155:MEDLINE(R)

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09142587 PMID: 1309513

**Nucleotide sequence of the lecithinase operon of *Listeria monocytogenes* and possible role of lecithinase in cell-to-cell spread.**

Vazquez-Boland J A; Kocks C; Dramsi S; Ohayon H; Geoffroy C; Mengaud J; Cossart P

Unite de Genie Microbiologique, Institut Pasteur, Paris, France.

Infection and immunity (UNITED STATES) Jan 1992, 60 (1) p219-30,

ISSN 0019-9567 Journal Code: 0246127

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Subfile: INDEX MEDICUS

The lecithinase gene of the intracellular pathogen *Listeria monocytogenes*, *plcB*, was identified in a 5,648-bp DNA fragment which expressed lecithinase activity when cloned into *Escherichia coli*. This fragment is located immediately downstream of the previously identified gene *mpl* (*prtA*). It contains five open reading frames, named *actA*, *plcB*, and *ORFX*, *-Y*, and *-Z*, which, together with *mpl*, form an operon, since a 5.7-kb-long **transcript** originates from a promoter located upstream of *mpl* (J. Mengaud, C. Geoffroy, and P. Cossart, *Infect. Immun.* 59:1043-1049, 1991). A second promoter was detected in front of *actA* which encodes a putative membrane protein containing a region of internal repeats. *plcB* encodes the lecithinase, a predicted 289-amino-acid protein homologous to the phosphatidylcholine-specific phospholipases C of *Bacillus cereus* and *Clostridium perfringens* (alpha-toxin). *plcB* mutants produce only small plaques on fibroblast monolayers, and an electron microscopic analysis of

infected macrophages suggests that lecithinase is involved in the lysis of the two-membrane vacuoles that surround the bacteria after cell-to-cell spread. On the opposite DNA strand, downstream of the operon, three more open reading frames, *ldh*, *ORFA*, and *ORFB*, were found. The deduced amino acid sequence of the first one is homologous to lactate dehydrogenases. Low-stringency Southern hybridization experiments suggest that these three open reading frames lie outside of the *L. monocytogenes* virulence region: *mpl* and *actA* were specific for *L. monocytogenes*, sequences hybridizing to *plcB* were detected in *L. ivanovii* and *L. seeligeri*, and sequences hybridizing to *ORFX*, *-Y*, and *-Z* were found in *L. innocua*. In contrast to this, sequences hybridizing to *ldh* or *ORFB* were detected in all *Listeria* species (including the nonpathogenic ones).

Tags: Comparative Study; In Vitro; Support, Non-U.S. Gov't

Descriptors: \**Listeria monocytogenes*--enzymology--EN; \*Operon--genetics--GE; \*Phospholipases--genetics--GE; Amino Acid Sequence; Animals; Bacterial Outer Membrane Proteins--genetics--GE; Base Sequence; Blotting, Southern; Cloning, Molecular; DNA Transposable Elements; *Listeria monocytogenes*--pathogenicity--PY; Mice; Microscopy, Electron; Molecular Sequence Data; Phospholipases--physiology--PH; Plaque Assay; Promoter Regions (Genetics)--genetics--GE; Restriction Mapping; Sequence Homology, Nucleic Acid; Virulence--genetics--GE

Molecular Sequence Databank No.: GENBANK/M63610; GENBANK/M63611; GENBANK/M63612; GENBANK/M63613; GENBANK/M63614; GENBANK/M63615; GENBANK/M63616; GENBANK/M63617; GENBANK/M82881; GENBANK/X63185

CAS Registry No.: 0 (Bacterial Outer Membrane Proteins); 0 (DNA Transposable Elements)

Enzyme No.: EC 3.1.- (Phospholipases)

Gene Symbol: *-y*; *-z*; *ORFX*; *actA*; *hyl*; *ldh*; *mpl*; *plcB*; *prtA*

Record Date Created: 19920212

Record Date Completed: 19920212

10/9/22 (Item 22 from file: 34)

DIALOG(R) File 34:SciSearch(R) Cited Ref Sci

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01712426 Genuine Article#: HV090 Number of References: 39

Title: PURIFICATION AND CHARACTERIZATION OF AN ADP-RIBOSYLTRANSFERASE PRODUCED BY CLOSTRIDIUM-LIMOSUM

Author(s): JUST I; MOHR C; SCHALLEHN G; MENARD L; DIDSBURY JR; VANDEKERCKHOVE J; VANDAMME J; AKTORIES K

Corporate Source: UNIV SAARLAND, INST PHARMACOL & TOXIKOL/W-6650 HOMBURG//GERMANY//; UNIV SAARLAND, INST PHARMACOL & TOXIKOL/W-6650 HOMBURG//GERMANY//; UNIV BONN, INST MED MIKROBIOL & IMMUNOL/W-5300 BONN//GERMANY//; LAB FYSIOL SCHEIKUNDE/B-9000 GHENT//BELGIUM//; DUKE UNIV, MED CTR, DEPT MED/DURHAM//NC/27710

Journal: JOURNAL OF BIOLOGICAL CHEMISTRY, 1992, V267, N15 (MAY 25), P 10274-10280

Language: ENGLISH Document Type: ARTICLE

Geographic Location: GERMANY; BELGIUM; USA

Subfile: SciSearch; CC LIFE--Current Contents, Life Sciences

Journal Subject Category: BIOCHEMISTRY & MOLECULAR BIOLOGY

Abstract: We purified a novel ADP-ribosyltransferase produced by a *Clostridium limosum* strain isolated from a lung abscess and compared the exoenzyme with *Clostridium botulinum* ADP-ribosyltransferase C3. The *C. limosum* exoenzyme has a molecular weight of about 25,000 and a *pI* of 10.3. The specific activity of the ADP-ribosyltransferase is 3.1 nmol/mg/min with a *K<sub>m</sub>* for NAD of 0.3- $\mu$ M. Partial amino acid sequence analysis of the tryptic peptides revealed about 70% homology with C3. The novel exoenzyme modifies selectively the small GTP-binding proteins of the rho family in human platelet membranes presumably at the same amino acid (asparagine 41) as known for C3. Recombinant rhoA and rhoB serve as substrates for C3 and the *C. limosum* exoenzyme. Whereas recombinant rac1 protein is only marginally ADP-ribosylated by C3 or by the *C. limosum* exoenzyme in the absence of detergent, in the presence of 0.01% sodium dodecyl sulfate rac1 is modified by C3 but not by the *C. limosum* exoenzyme. Recombinant CDC42Hs protein is a poor

substrate for C. limosum exoenzyme and is even less modified by C3.

The C. limosum exoenzyme is auto-ADP-ribosylated in the presence of 0.01% sodium dodecyl sulfate by forming an ADP-ribose protein bond highly stable toward hydroxylamine. The data indicate that ADP-ribosylation of small GTP-binding proteins of the rho family is not unique to C. botulinum C3 ADP-ribosyltransferase but is also catalyzed by a C3-related exoenzyme from C. limosum.

Identifiers--KeyWords Plus: RHO-GENE-PRODUCT; PERFRINGENS IOTA TOXIN; GTP-BINDING PROTEINS; BOTULINUM ADP-RIBOSYLTRANSFERASE-C3; POLYACRYLAMIDE GELS; ESCHERICHIA-COLI; SKELETAL-MUSCLE; ACTIN; RIBOSYLATION; SUBSTRATE

Research Fronts: 90-3110 003 (IDENTIFICATION OF FRAGMENTS; CORTICOSTEROIDS INCREASE LIPOCORTIN-I; RAS ADENYLATE-CYCLASE PATHWAY; HEAT-SHOCK PROTEIN HSP70 FAMILY)

90-3780 001 (PERTUSSIS TOXIN; VASOPRESSIN SENSITIVE ADENYLATE-CYCLASE; SIGNAL TRANSDUCTION MECHANISM)

90-6257 001 (ADENOVIRUS-2 MAJOR LATE PROMOTER; INVITRO **TRANSCRIPTION** ; YEAST PROTEIN; UPSTREAM ELEMENT FACTOR; RECOGNITION OF DNA; ACTIVE NF-KAPPA-B)

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?t s10/3,kwic/2

>>>KWIC option is not available in file(s): 399

10/3,KWIC/2 (Item 2 from file: 399)

DIALOG(R)File 399:CA SEARCH(R)

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123307419 CA: 123(23)307419r JOURNAL

The construction of a reporter system and use for the investigation of  
*Clostridium perfringens* gene expression

AUTHOR(S): Bullifent, Helen L.; Moir, Anne; Titball, Richard W.

LOCATION: Chemical and Biological Defence Establishment, Porton Down,  
Salisbury, UK, SP4 0JQ

JOURNAL: FEMS Microbiol. Lett. DATE: 1995 VOLUME: 131 NUMBER: 1

PAGES: 99-105 CODEN: FMLED7 ISSN: 0378-1097 LANGUAGE: English

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\$6.50 Estimated cost File434  
\$0.06 0.013 DialUnits File444  
\$0.06 Estimated cost File444  
\$0.09 0.013 DialUnits File467  
\$0.09 Estimated cost File467  
OneSearch, 26 files, 0.671 DialUnits FileOS  
\$0.24 TELNET  
\$63.36 Estimated cost this search  
\$63.36 Estimated total session cost 0.671 DialUnits

### Status: Signed Off. (1 minutes)

03238985 Genuine Article#: NP484 Number of References: 80

**Title: IDENTIFICATION AND MOLECULAR ANALYSIS OF A LOCUS THAT REGULATES  
EXTRACELLULAR TOXIN PRODUCTION IN CLOSTRIDIUM - PERFRINGENS**

Author(s): LYRISTIS M; BRYANT AE; SLOAN J; AWAD MM; NISBET IT; STEVENS DL;  
ROOD JI

Corporate Source: MONASH UNIV, DEPT MICROBIOL/CLAYTON/VIC 3168/AUSTRALIA/;  
MONASH UNIV, DEPT MICROBIOL/CLAYTON/VIC 3168/AUSTRALIA/; VET ADM MED  
CTR, INFECT DIS RES UNIT/BOISE//ID/83702; UNIV WASHINGTON, SCH MED, DEPT  
MED/SEATTLE//WA/98195; COMMONWEALTH SERUM LABS/PARKVILLE/VIC  
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Journal: MOLECULAR MICROBIOLOGY, 1994, V12, N5 (JUN), P761-777

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**Abstract:** The anaerobic bacterium *Clostridium perfringens* mediates clostridial myonecrosis, or gas gangrene, by producing a number of extracellular toxins and enzymes. Transposon mutagenesis with Tn916 was used to isolate a pleiotropic mutant of *C. perfringens* that produced reduced levels of phospholipase C, protease and sialidase, and did not produce any detectable perfringolysin O activity. Southern hybridization revealed that a single copy of Tn916 had inserted into a 2.7 kb HindIII fragment in the *C. perfringens* chromosome. A 4.3kb PstI fragment, which spanned the Tn916 insertion site, was cloned from the wild-type strain. When subcloned into a shuttle vector and introduced into *C. perfringens* this fragment was able to complement the Tn916-derived mutation. Transformation of the mutant with **plasmids** containing the 2.7kb HindIII fragment, or the 4.3kb PstI fragment, resulted in toxin and enzyme levels greater than or equal to those of the wild-type strain. The PstI fragment was sequenced and found to potentially encode seven open reading frames, two of which appeared to be arranged in an operon and shared sequence similarity with members of two-component signal transduction systems. The putative *virR* gene encoded a protein with a deduced molecular weight of 30140, and with sequence similarity to activators in the response regulator family of proteins. The next gene, *virS*, into which Tn916 had inserted, was predicted to encode a membrane-spanning protein with a deduced molecular weight of 51274. The putative *VirS* protein had sequence similarity to sensor proteins and also contained a histidine residue highly conserved in the histidine protein kinase family of sensor proteins. Virulence studies carried out using a mouse model implicated the *virS* gene in the pathogenesis of histotoxic *C. perfringens* infections. It was concluded that a two-component sensor regulator system that activated the expression of a number of extracellular toxins and enzymes involved in virulence had been cloned and sequenced. A model that described the regulation of extracellular toxin production in *C. perfringens* was constructed.

**Identifiers--KeyWords Plus:** PHOSPHOLIPASE-C GENE; TRANSFERABLE TETRACYCLINE RESISTANCE; EXPERIMENTAL GAS-GANGRENE; O THETA-TOXIN; ESCHERICHIA-COLI; **NUCLEOTIDE** -SEQUENCE; ALPHA-TOXIN; AGROBACTERIUM-TUMEFACIENS; SIGNAL TRANSDUCTION; PHOSPHATE REGULON